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CONTENTS

Vol. XXV, Part I

(March 1955)

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Original articles

	Page
EFFECT OF CONTINUOUS MANURING WITH ARTIFICIAL FERTILIZERS ON <i>Azotobacter</i> AND SOIL FERTILITY	B. N. Pramanik and A. N. Misra 1
A STUDY OF SOME OF THE PROPERTIES OF RED AND LATERITE SOILS OF BIHAR	Awadh Bihari Lall 15
UTILIZATION OF JUNGLE DRIED BONES	P. C. Juneja, K. C. Gulati and S. P. Raychandhuri 29
RATOONING OF SUGARCANE AS INFLUENCED BY THE TIME OF HARVESTING PLANT CANE (WITH PLATE I)	K. R. Tandon, G. P. Kapoor and G. N. Misra 31
CYTOGENETICAL INVESTIGATIONS ON GROUNDNUTS	C. N. Babu 41
I. THE SOMATIC CHROMOSOMES (WITH ONE TEXT-FIGURE)	
A CHROMOSOMAL CHIMERA IN <i>S. Halpense</i> (LINN.) (WITH PLATES II TO IV)	V. S. Raman and N. Krishna-swamy 47
INVESTIGATION ON THE PHYSIOLOGY OF GROWTH AND DEVELOPMENT OF <i>Crotalaria juncea</i> L. C. 12	J. C. Sen Gupta and Sumitra Talukdar 51
STUDIES ON INDIAN CLOVER (<i>Senji</i>) IN THE PUNJAB	H. C. Malik 67
A PRELIMINARY NOTE ON THE ESSENTIAL OIL BEARING PLANTS OF KASHMIR (PART V)	K. L. Handa, L. D. Kapoor and O. N. Channa 73
RAPID ESTIMATION OF YIELD AND IODINE VALUE OF OILS IN SMALL SAMPLES OF OILSEEDS	A. R. S. Kartha, A. S. Sethi and K. C. Gulati 79

Review

METHODS OF SURVEYING AND MEASURING VEGETATION	85
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ORIGINAL ARTICLES

EFFECT OF CONTINUOUS MANURING WITH ARTIFICIAL FERTILIZERS ON *AZOTOBACTER* AND SOIL FERTILITY

By B. N. PRAMANIK and A. N. MISRA, Main Sugarcane Research Station, Shahjahanpur

(Received for publication on 28 November 1953)

THE problems of soil fertility have been studied from varied aspects and it has been recognised that Mitcherlich's [Willcox, 1930] extension of Liebig's theory would hold good only when the soil is in a state of optimum biological activity. The use of fertilizers is no doubt, necessary for reaping high yields but the soil, as the seat of biological activity, also deserves equal consideration; in addition to humus, pH, supply of oxygen and available nutrients too play a very important part both in crop production and biological activity. The micro-organisms are sensitive to small differences of environmental conditions and, therefore, while practising the continuous application of nitrogenous, potassic and phosphatic fertilizers, due consideration must be given to the soil micro-organisms which are responsible for making the nutrients available for plant growth. Various attempts have been made in the past, some of them with remarkable success, to correlate the number of soil micro-organisms with soil fertility, it would, therefore, be of interest to study the population of micro-organisms in plots subjected to continuous application of nitrogenous, phosphatic and potassic fertilizers. Since biological processes are partly responsible for building soil fertility, the methods of microbiological analysis may be taken advantage of in these studies recognising the fact that soil micro-organisms have the same requirements for growth as plants. According to Berjerinck [1901], the number of *Azotobacter* in soil runs parallel with soil fertility; and accordingly *Azotobacter* activities may serve also as an index of soil fertility. *Azotobacter* sp. resists drying readily and is sensitive to supplies of available carbohydrates and phosphorus in the soil, and soil activity. Advantage of these facts has been taken by many workers in finding out the manurial requirements of soils.

Direct observation on soil plaque has been carried out by Winogradsky [1926] and the technique of plaque moulee (the kneaded soil method) devised by him was later developed by others. He and Tiemiecka [1928] have developed a method for directly studying *Azotobacter* of soil with a view to establishing its relation with growing plant in natural environments. Ziemiecka [1932] showed that nitrogenous manures had harmful effect on *Azotobacter* sp. and phosphate in combination with potash responded better than when they were applied separately. Guittonean [1929] obtained good correlation between *Azotobacter* growth on soil plaque and field conditions. Konishi and Tsuge [1933], Vandecavve and Anderson [1933] and Bortel [1937] observed that the number of *Azotobacter* increases with the application of phosphorus, zinc and molybdenum to the soil. Hill [1917] found that nitrogenous compounds generally depress nitrogen fixation in soil whereas Schneider

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[1931] observed that the application of NaNO_3 and urea favoured the growth of *Azotobacter chroococcum*, while continuous dressing of sulphate of ammonia retarded its development. Jones [1932] revealed that the growth of *Azotobacter* in soil plaque bears a satisfactory correlation with crop yield. Joshi and Ram Iyer [1933] have tried Winogradsky's technique modified by Sackett and Stewart [1931] for Indian soils and observed that the method is of value for determining the availability of different phosphatic fertilizers, for testing the activity of soils, and for finding their lime requirements. Stockli [1946] found that *Azotobacter chroococcum* numbers are positively correlated to both calcium carbonate and available phosphorus in soil. Desai [1946] adopted the *Azotobacter* plaque method in assessing the fertility of Pusa soils. The *Azotobacter* plaque test being comparatively simple and fairly accurate has been adopted here for studying the effect of continuous application of sulphate of ammonia, superphosphate and potassium sulphate on *Azotobacter chroococcum* of sugarcane soils.

MATERIALS AND METHOD

The Complete Nutrient Experiment at this Research Station was selected for studying this problem. This experiment was laid out in 1935 and consists of 27 manurial treatment combinations, at three levels (0, 100 and 200 lb.) of nitrogen, three levels (0, 75 and 150 lb.) of phosphate and three levels (0, 75 and 150 lb.) of potash. The layout adopted is the split-plot design, with levels of N in main plots and with four replications. The experiment is being conducted on two fields H3 and H4 in alternate years following the rotation cane-fallow-cane.

The results in regard to the cane yield and juice quality show no response to P_2O_5 or K_2O and the interactions between these three main manurial treatments, N, P, K., are also not significant. On an average, the response of 100 lb. nitrogen as sulphate of ammonia is 288 maunds whereas the additional 100 lb. nitrogen increased the cane yield by only 40 maunds per acre. During the first few years high yields of cane were obtained while from 1943 the cane yields started decreasing and the response to nitrogen has also diminished. The yields from plots treated with sulphate of ammonia alone, have shown greater fall than those from unmanured (control) plots. Nitrogen application lowers the quality of juice whereas P_2O_5 and K_2O treatments show no such effects. The yields of cane under the different manurial treatments under examination are given in Appendix I. The methods adopted for the various chemical and microbiological determinations are given below :

The pH estimations were carried out by means of Hellige's comparator with phenolred and chlorophenol as internal indicators.

The nitrogen estimations were done by usual Kjeldahl's modified method.

Ammonia was estimated by the Mclean Robinson method [1924] and the nitrite by Griess Ilosvays method.

Nitrate estimates were done by Harper's [1924] phenol disulphonic acid method.

The total population of micro-organisms was determined in Thorton's agar medium. The method used for suspension, disintegration, dilution, plating and incubation was after Russell [1932].

Twenty-four soils pertaining to the four replicates of six treatment combinations, viz. N1P1K1, N1P1K3, N1P3K1, N3P1K1, N1P3K3 and N3P3K3 were studied in the first year and during the second year, the three additional treatments, viz. N2P1K1, N3P3K1 and N3P1K3, were also included thus making (24+36) 60 samples in all for *Azotobacter* plaque test studies. Composite soil samples were taken for chemical analysis.

DATA AND DISCUSSION

For *Azotobacter* studies, soil samples 0-9 in. depth were taken from each of the four replicates before the application of manures. Soil samples were taken during both the years 1947-48 and 1948-49 from the fields H₃ and H₄ of the Research Station in which the experiment is being carried out. For each of the manurial treatments (N, P and K), 50 gm. soil samples were taken and to each 0.5 gm. of mannite was added. Four such 50 gm. portions of the individual sample, termed as A, B, C, D were treated with the following for studying the response of additional phosphate and potash. :—

A—No further salt was added (Control).

B—5 c.c. of 3 per cent solution of potash sulphate added.

C—5 c.c. of 6 per cent solution of disodium hydrogen phosphate added.

D—5 c.c. of 3 per cent solution of dipotassium hydrogen phosphate added.

While preparing the soil plaque, equal quantities of sterile distilled water were added to all the treatments. The soil plaques were incubated for 48 hours at 30°C. in a moist chamber. The bead-like, white, glistening colonies of *Azotobacter* per sq. cm. were counted at three different places over the plaque surface in order to obtain the average counts. The results of *Azotobacter* counts of all the twenty-four plots for the year 1947-48 and thirty-six plots for the year 1948-49 are given in the Appendices II(a) and II(b) and the summary of results in Tables I and II.

TABLE I(a)

Summary of results of *Azotobacter* plaque studies 1947-48 as per reagents used (Field H₄)

	K ₂ HPO ₄	K ₂ SO ₄	Na ₂ HPO ₄	Control
Reagents Mean No. of <i>Azotobacter</i> colonies	19.48	5.75	5.55	4.21 (Sig. at P=0.01)

TABLE I(b)

Summary of results of *Azotobacter* plaque studies 1947-48 as per field treatments (Field H₄)

	N1P3K1	N1P3K3	N3P3K3	N1P1K3	N1P1K1	N3P1K1
Field Treatments Mean No. of <i>Azotobacter</i> colonies	15.10	12.10	8.99	6.21	4.30	3.62
(Sig. at P=0.05)						

TABLE II(a)

Summary of results of *Azotobacter* plaque studies 1948-49 as per reagents used (Field H₃)

	K ₂ HPO ₄	Na ₂ HPO ₄	K ₂ SO ₄	Control
Reagents Mean No. of <i>Azotobacter</i> colonies	29.53	5.36	3.70	3.62 (Sig. at P=0.01)

TABLE II(b)

Summary of results of *Azotobacter* plaque studies 1948-49 as per field treatments (Field H₃)

	* N1P3K1	N1P3K3	N1P1K1	N1P1K3	N3P3K3	N3P ₂ K1	N3P1K1	N3P1K3
Field Treatments Mean No. of <i>Azotobacter</i> colonies	21.59	18.95	12.82	7.85	5.70	5.31	3.34	1.57
	(Sig. at P=0.05)							

* N1=0 lb. N per acre

• P1=0 lb. P₂O₅ per acre

K1=0 lb. K₂O per acre

N3=200 lb. N. per acre

P3=150 lb. P₂O₅ per acre

K3=150 lb. K₂O per acre

Results obtained during both the years indicate :

- That the continuous application of sulphate of ammonia in fields has produced a depressing effect on *Azotobacter* growth.
- That on the average, phosphate manured plots (P3) have resulted in significantly more *Azotobacter* colonies per sq. cm. than the rest. It is, therefore, indicated that phosphate treatment increases nitrogen fixing capacity of the soil and its fertility (*vide* Appendix III).
- That amongst the reagents added in the laboratory to these soils for further testing the response to additional phosphate and potash, K₂HPO₄, gave significantly higher average count. There were no significant differences among K₂SO₄, Na₂HPO₄ and control.
- That the difference in response to the two phosphatic salts, viz. Na₂HPO₄ and K₂HPO₄ on the growth of *Azotobacter* shows a capacity for selection by the *Azotobacter* strains, indicating thereby the relative availability of phosphates to the micro-organisms in different phosphatic fertilizers.

To test the fertility status of these soils after receiving continued application of these fertilizers, the population of micro-organisms, pH, total nitrogen, available nitrogen and organic carbon were determined for both the fields (H₄ and H₃). The results obtained are given in Table III. The soils of the four replicates of each treatment were thoroughly mixed and representative composite samples taken for the above determinations. The bacterial counts on Thorton's agar medium were made after incubating for seven days at 30°C.

TABLE III

Bacterial population, pH, total and available nitrogen and organic carbon in the two fields (H_4 and H_3)

Field	Treatments*	Total bacterial population on Thorton's agar medium per gm. (Millions)	pH	Total nitrogen	Carbon	C/N ratio	Nitrification	
							Ammonia and Nitrite	Nitrates ($\text{NO}_3\text{-N}$) in mg. per 100 gm. soil
H_4	N1P1K1	6.75	7.4	0.0406	0.209	5.15	Trace	0.21
	N1P1K3	4.00	7.4	0.0420	0.232	5.52	Trace	0.22
	N1P3K1	8.10	7.4	0.0434	0.213	4.91	Trace	0.31
	N1P3K3	8.75	7.4	0.0374	0.243	6.43	Trace	0.17
	N3P1K1	3.00	6.7	0.0378	0.239	6.32	Trace	0.16
	N3P1K3	3.05	6.4	0.0392	0.217	5.54	Trace	0.19
	N3P3K1	5.65	6.7	0.0364	0.236	6.48	Trace	0.18
	N3P3K3	2.35	6.0	0.0420	0.201	4.79	Trace	0.17
	N2P1K1	6.15	6.8	0.0448	0.220	4.91	Trace	0.10
H_3	N1P1K1	4.50	7.2	0.0476	0.239	5.02	Trace	0.61
	N1P1K3	4.25	7.0	0.0490	0.239	4.88	Trace	0.82
	N1P3K1	4.75	7.1	0.0448	0.220	4.91	Trace	0.74
	N1P3K3	4.00	6.9	0.0378	0.255	6.75	Trace	0.88
	N3P1K1	1.75	6.3	0.0504	0.205	4.07	Trace	0.84
	N3P1K3	1.90	5.8	0.0476	0.228	4.79	Trace	0.60
	N3P3K1	2.50	6.4	0.0532	0.217	4.08	Trace	0.87
	N3P3K3	2.50	5.7	0.0574	0.217	3.78	Trace	0.76
	N2P1K1	3.90	6.8	0.0490	0.194	3.96	Trace	0.61

* N1=0 lb. N per acre

P1=0 lb. P_2O_5 per acreK1=0 lb. K_2O per acre

N2=100 lb. N per acre

P3=150 lb. P_2O_5 per acreK3=150 lb. K_2O per acre

N3=200 lb. N per acre

The data indicate that the total bacterial population has been depressed in plots treated continuously with large doses of sulphate of ammonia. It is interesting to note that this depressing effect is less in evidence when the dosage of sulphate of ammonia is reduced from (200 lb. N per acre) to N_2 (100 lb. N) level. The average rate of fall in yield of cane from the year of commencement of the experiment up to date is greater in N_3 plots than in others. This fall in yield and the diminishing response to nitrogen noticed specially from the year 1943 onwards may be ascribed to the low bacterial activity of the corresponding soils. The application of potash appears to be unfavourable for microbiological population and has shown no response to crop yields when applied even at 150 lb. K_2O per acre. The beneficial effects of

phosphates have not resulted in higher yields presumably as these were applied as top dressings and thereby got immobilised in the surface soil.

It may be noted that pH value of the soil has been appreciably reduced and the soil rendered acidic due to the continuous application of sulphate of ammonia (N3 treatment). From the analytical data dealing with total nitrogen, nitrate nitrogen and carbon it is quite evident that the continued cropping with sugarcane has appreciably reduced the C/N ratio, total carbon and available nitrogen of the soils.

To study the nitrogen fixing capacity in the presence of energy giving carbohydrate material (Mannite) of the soils of the plots under various manurial treatments of the Complete Nutrient Experiment, a series of experiments were started in the laboratory. The soil samples (0-9 in.) from various plots of the field (H_4) were taken after the application of manures in the year (1949-50) and composite air dry soil sample was used for the following studies. To 20 gm. of soil sample representing each of the different treatments, 2 per cent mannite was added and the soils in duplicate were incubated in 250 c.c. Erlenmeyer flasks for 75 days at 30°C. Water equal to one-third saturation capacity was added and maintained throughout the entire period of incubation. The final gain of nitrogen was estimated at the end of the period by modified Kjeldahl's method. In another series of these flasks, same quantity of soil in duplicate was taken and 2 per cent mannite added in combination with 0.5 per cent K_2HPO_4 per flask. Total nitrogen was estimated as before after incubating them at 30°C. at one-third moisture-holding capacity for a period of 75 days. The results obtained are given in Appendix III and the summary of results in Tables IV and V.

TABLE IV
Summary of results of nitrogen fixation as per field treatments
(milligrams per 100 gm. soil)

	*P1K1	P3K1	P1K3	P3K3	Total	Average
N1	11.9	13.2	4.6	10.9	40.6	10.2
N2	3.9	4.9	2.1	6.0	16.9	4.2
N3	7.4	9.1	2.8	9.8	29.1	7.3
Total	23.2	27.2	9.5	26.7		
Average	7.7	9.1	3.2	8.9		
Mean of K1		8.4	—			
Mean of K3		—		6.1		

*P1=0 lb. P_2O_5 per acre
P3=150 lb. P_2O_5 per acre

K1=0 lb. K_2O per acre
K3=150 lb. K_2O per acre

It may be seen from Table IV that there is a significant reduction of nitrogen fixation in the cases of soils treated with nitrogen and potash. The soils treated with P_2O_5 (P3, P3K3, N3P3K3) have invariably given relatively more nitrogen fixation.

TABLE V

*Summary of results of nitrogen fixation as per additional treatment of K_2HPO_4 ,
(milligrams per 100 gm. soil)*

	PIK1	P3K1	PIK3	P3K3	Total	Average
N1 (0 lb. N)	24.5	38.4	10.5	17.9	91.3	22.8
N2 (100 lb. N)	22.8	12.3	5.6	7.4	48.1	12.0
N3 (200 lb. N)	11.2	11.6	5.6	14.7	43.1	10.8
Total	58.5	62.3	21.7	40.0		
Average	19.5	20.8	7.2	13.3		
Mean of K1(0 lb. K_2O)		20.2		—		
Mean of K3 (150 lb. K_2O)		—		10.3		

There is further confirmation of the findings given in Table IV from the summary results given in Table V, viz. soils treated with sulphate of ammonia have significantly lower capacity for nitrogen fixation and soils treated with potassium also behave in a similar manner. It may be seen that addition of K_2HPO_4 has resulted in an increased nitrogen fixation thereby confirming the earlier findings regarding the usefulness of phosphatic manuring for improving the soil fertility and also the detrimental effect of continuous application of sulphate of ammonia on the nitrogen fixing capacity of the soils.

Continuous application of sulphate of ammonia as revealed by the data given in the foregoing pages, shows considerable decrease of pH of soils. This is an already well-known fact that sulphate of ammonia mobilizes lime and increases soil acidity. In addition the soils show a considerable reduction in total bacterial counts as well as loss of recouperation of nitrogen through the fixation of atmospheric nitrogen. While there is very little reduction in pH values of soils between applications of 100 lb. and 200 lb. N per acre, there is considerable reduction in total bacterial counts. This might have been caused by the high concentration of soluble nutrients bringing about plasmolysis of bacteria. It would, therefore, not be justifiable to apply highly concentrated soluble nutrients in large quantities as that would ultimately result in low biological activity.

Continuous application of superphosphate upto 150 lb. P_2O_5 per acre has not resulted in either a depression of bacterial counts or reduction of the population of *Azotobacter*. It may be conceded that interaction of soil and superphosphate reduces temporarily its availability but does not lead to high concentration of soil solution, to the extent of being detrimental to microbiological population. On the other hand, presence of sufficient amount of phosphorus in the substratum provides conditions for multiplication of bacteria. The effect of potassic fertilizers, however, depresses the nitrogen fixing capacity of the soil and also does not lead to any increase in the bacterial counts. Iversen and Petersen [1948] came to a like conclusion in respect of application of high doses of potash but their observations with regard to superphosphate have not been confirmed in our studies.

Where nutrients like K_2HPO_4 , Na_2HPO_4 and K_2SO_4 are added in the laboratory to these soils, and *Azotobacter* counts made by the plaque method, K_2SO_4 alone shows a depressing effect. Between K_2HPO_4 and Na_2HPO_4 , it is the former that has led to more favourable conditions for *Azotobacter* growth. It appears that *Azotobacter* prefers P in combination with K. It is well-known that K and Na ions behave differently when added to soils. Sodium ions create undesirable physical conditions in soil whereas potassium ions behave in a manner similar to that of lime. This may be the underlying reason for the differential behaviour of the two chemicals. Nevertheless, potassium salts alone have on the whole a depressing effect on *Azotobacter*. It may be seen from these investigations that continuous application of sulphate of ammonia without superphosphate causes adverse conditions for proper biological activity. For the maintenance of soil fertility under intense cropping conditions, it is desirable to supply sufficient energy-giving material for bacterial life. The present conditions of these soils are such that not only organic matter should be applied but also sufficient lime must be incorporated into these soils especially in N_2 and N_3 soils, to improve the conditions for microbiological activity. (Since this note was prepared *sanai* green manuring has been introduced in this experiment and later results have shown great improvement in the fertility status of the fields.)

SUMMARY

(1) Soils from six treatments (H4) in the first year and nine treatments (H3) in the second year of the four replicates of the Complete Nutrient Experiment at Shahjahanpur Research Station, in which nitrogen, phosphorus and potash are applied continuously in three doses during the past seven years, have been examined chemically and biologically.

(2) It has been observed that continuous application of concentrated nitrogenous fertilizers $(NH_4)_2SO_4$ decreases the total number of soil micro-organisms and hampers nitrogen fixation by creating unfavourable soil reactions.

(3) Application of phosphatic fertilizers creates favourable conditions for the growth of *Azotobacter* sp. and other soil micro-organisms. It increases the nitrogen recuperative power of the soil by increased bacterial fixation of atmospheric nitrogen. The beneficial effect of the phosphates has not resulted in higher yields presumably

as these were applied as top dressings and thereby got immobilised in the surface soil.

(4) The supply of potassic fertilizer (K_2SO_4) depresses nitrogen fixing capacity of these soils and thus hinders soil recouperation. Hence it does not lead to increased yields.

(5) The *Azotobacter* plaque test may be utilised for comparing the availability of various phosphatic fertilizers.

(6) Continuous cropping of sugarcane in the absence of organic manuring both with and without application of phosphatic fertilizers exhausts the fertility of the soil and lowers the C/N ratio.

(7) The steeper decline in cane yields from plots treated with sulphate of ammonia may be ascribed to markedly low bacterial activity in those soils.

(8) The low fertility level of the soil can be improved through increased nitrogen recuperative capacity of the soil micro-organisms by the application of organic matter and phosphatic fertilizers at a depth of about four to six inches in the soil.

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APPENDIX I

Average cane yield in maunds per acre from 1935-36 to 1948-49

No.	Treatments	Variety						
		Co. 313	Co. 421	Co. 381	Co. 421	Co. 421	Co. 421	Co. 421
		1935-36	1937-38	1939-40	1941-42	1943-44	1945-46	1947-48
				<i>Field No. H4</i>				
1	N1P1K1*	591-90	371-05	599-25	282-04	424-10	463-93	383-70
2	N1P1K3	539-50	322-62	546-99	237-40	416-20	384-11	387-40
3	N1P3K1	596-70	357-81	560-92	264-57	384-30	367-11	348-10
4	N1P3K3	552-40	360-59	567-89	274-92	416-20	460-30	423-90
5	N3P1K1	861-50	783-20	891-90	687-21	628-80	577-99	497-80
6	N3P1K3	886-40	841-39	871-00	727-45	688-70	634-67	515-60
7	N3P3K1	912-00	820-48	905-84	749-53	695-90	599-75	513-70
8	N3P3K3	894-20	808-06	898-87	740-27	710-20	619-48	546-50
9	N2P1K1	899-30	795-75	864-03	578-48	644-90	486-38	490-30

No.	Treatments	Variety						
		Co. 313	Co. 421	Co. 331	Co. 421	Co. 421	Co. 421	Co. 421
		1936-37	1938-39	1940-41	1942-43	1944-45	1946-47	1948-49
				<i>Field No. H3</i>				
1	N1P1K1	350-58	550-36	374-08	458-92	400-97	263-90	292-9
2	N1P1K3	405-28	568-34	421-60	490-33	404-01	299-40	280-7
3	N1P3K1	390-09	575-63	404-42	495-95	472-09	321-90	277-0
4	N1P3K3	382-19	514-39	367-56	454-77	444-48	264-80	260-1
5	N3P1K1	791-68	845-82	612-90	1037-75	777-37	495-00	506-2
6	N3P1K3	787-73	913-67	534-16	1083-60	783-46	497-80	549-3
7	N3P3K1	771-56	862-52	521-64	1053-65	749-30	535-30	571-7
8	N3P3K3	793-47	899-28	536-85	987-23	787-20	509-00	562-4
9	N2P1K1	626-09	895-68	502-85	967-57	700-17	448-20	483-8

*N1=0 lb. N per acre
N2=100 lb. N per acre
N3=200 lb. N per acre

P1=0 lb. P₂O₅ per acre
P3=150 lb. P₂O₅ per acre

K1=0 lb. K₂O per acre
K3=150 lb. K₂O per acre

APPENDIX II(a)
Azotobacter colony counts on soil-plaques during 1947-48

Soils (Field No. H4) before application of manures	Replication	Control, i.e., soils taken from field from each treatment per 1 sq. cm.	Treated with		
			K ₂ SO ₄ 1 sq. cm.	Na ₂ HPO ₄ 1 sq. cm.	K ₂ HPO ₄ 1 sq. cm.
I.	N1P1K1	nil	2	11	14
	N1P1K3	nil	nil	5	18
	N1P3K1	11	14	22	23
	N3P1K1	6	2	10	13
	N1P3K3	15	10	6	21
	N3P3K3	11	10	10	15
II.	N1P1K1	nil	4	3	15
	N1P1K3	nil	nil	3	16
	N1P3K1	6	12	12	20
	N3P1K1	2	3	nil	7
	N1P3K3	12	3	12	24
	N3P3K3	16	19	18	21
III.	N1P1K1	7	14	20	32
	N1P1K3	nil	nil	11	29
	N1P3K1	30	34	19	35
	N3P1K1	nil	9	nil	24
	N1P3K3	23	33	12	28
	N3P3K3	15	nil	nil	15
IV.	N1P1K1	nil	3	nil	10
	N1P1K3	nil	9	1	21
	N1P3K1	4	4	1	24
	N3P1K1	nil	3	nil	3
	N1P3K3	nil	nil	4	29
	N3P3K3	nil	2	nil	29

APPENDIX II(b)

Azotobacter colony counts on soil plaques during 1948-49

No.	Treatments (Field H3) before application of manures during the year 1948-49	Control, i.e., soils taken as per treatments	Treated with		
			K ₂ SO ₄	Na ₂ HP0 ₄	K ₂ HP0 ₄
1	N1P1K1	5	1	7	54
2	N1P1K3	12	10	5	47
3	N1P3K1	22	26	31	53
4	N3P1K1	1	-nil-	-nil-	21
5	N1P3K3	17	32	12	33
6	N3P1K3	3	2	3	4
7	N3P3K1	10	17	8	18
8	N3P3K3	9	nil	6	22
9	N2P1K1	2	nil	1	15
1	N1P1K1	5	10	nil	165
2	N1P1K3	nil	nil	nil	60
3	N1P3K1	9	6	41	84
4	N3P1K1	1	nil	5	42
5	N1P3K3	22	40	46	124
6	N3P1K3	nil	2	nil	9
7	N3P3K1	2	nil	15	35
8	N3P3K3	5	23	nil	17
9	N2P1K1	nil	nil	3	14
1	N1P1K1	nil	nil	nil	30
2	N1P1K3	-nil-	-nil-	6	21
3	N1P3K1	9	5	5	23
4	N3P1K1	nil	nil	4	12
5	N1P3K3	4	9	nil	23
6	N3P1K3	nil	nil	nil	5
7	N3P3K1	nil	nil	nil	26
8	N3P3K3	nil	nil	18	23
9	N2P1K1	4	nil	nil	24
1	N1P1K1	12	14	18	38
2	N1P1K3	2	nil	5	49
3	N1P3K1	20	20	16	28
4	N3P1K1	nil	nil	nil	16
5	N1P3K3	3	nil	13	27
6	N3P1K3	nil	nil	nil	8
7	N3P3K1	nil	nil	nil	5
8	N3P3K3	nil	nil	4	6
9	N2P1K1	nil	nil	nil	18

APPENDIX III

Nitrogen fixation capacity in each of the manurial treatments with and without the addition of K_2HPO_4 (nitrogen in mg. per 100 gm. soil)

Field Treatments	Nitrogen fixation in flasks with 2 per cent mannite and 20 gm. soil incubated at room temperature for a period of 75 days					
	As per field treatments			With the addition of 0.5 per cent K_2HPO_4 per flask in the laboratory		
	Nitrogen at start	Nitrogen after 75 days	Increase of nitrogen in 75 days	Nitrogen after 75 days	Increase of nitrogen in 75 days	Increase of nitrogen due to K_2HPO_4 (Col. 6 - Col. 4)
1	2	3	4	5	6	7
N1P1K1	54.6	66.5	11.9	79.1	24.5	12.6
N1P1K3	49.3	52.9	4.6	58.8	10.5	5.9
N1P3K1	54.0	67.2	13.2	92.4	38.4	25.2
N1P3K3	50.4	61.8	10.9	68.3	17.9	7.0
N2P1K1	49.3	52.2	3.9	71.1	22.8	13.9
N2P1K3	49.7	51.8	2.1	55.3	5.6	3.5
N2P3K1	46.9	51.8	4.9	59.2	12.3	7.4
N2P3K3	50.4	56.4	6.0	57.8	7.4	1.4
N3P1K1	47.6	55.0	7.4	58.8	11.2	3.8
N3P1K3	56.7	59.5	2.8	62.3	5.6	2.8
N3P3K1	50.0	65.1	9.1	67.6	11.6	2.5
N3P3K3	56.7	66.5	9.8	71.4	14.7	4.9

A STUDY OF SOME OF THE PROPERTIES OF RED AND LATERITIC SOILS OF BIHAR

By AWADH BIHARI LALL, Indian Agricultural Research Institute, New Delhi

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(With two text-figures)

RED and laterite soils form two of the four major types of Indian soils, together covering approximately a third of the culturable land of the country. The red soils including red loam and yellow earths occupy a total area of about 200,000 square miles while 49,000 square miles comprise laterite and lateritic soils. These soils extend over a large part of Madras, Orissa, Mysore, south-eastern Bombay and the eastern parts of Hyderabad and Madhya Pradesh. They also occur in Chota Nagpur of Bihar, Bankura, Birbhum and Midnapur districts of West Bengal, Khasi and Jaintia Hills of Assam and also some parts of U. P. The red soils are porous and friable and are free from lime *kankar* and carbonates. The soluble salts are usually low. The soils are generally neutral to acid in reaction and are deficient in nitrogen, phosphoric acid, lime and organic matter. A characteristic feature of the laterites is the presence of indurated honey-combed mass which is developed from the separation of iron oxide in the form of nodules and gradual cementation of the latter. Such honey-combed structure is absent in red and lateritic soils.

A complete review of the existing literature of lateritic and red soils of India has been made by Raychaudhuri [1937, 1941 and 1943] and distinctive properties of these soils have been pointed out by him. Vauder Marwe [1948] considers that laterite and lateritic types soil formation require for their genesis a high precipitations, high temperature and efficient surface and internal drainage. These factors result in rapid and complete disintegration and decomposition of the parent rock and soil material. Silica thus released leaches down the profile while the sesquioxides separated out are fixed on the surface horizon. Very little accumulation of organic matter takes place in spite of abundance of vegetation. Preese (1949) has recently given a clear and precise definition of laterite. According to him a laterite is a 'Vesicular, concretionary, cellular, vermicular, clay-like, pisolitic or concrete-like mass consisting chiefly of ferric iron oxides, with or without mechanically entangled quartz and minor quantities of aluminium and manganese. It is of varying hardness but is usually easily shattered when struck a sharp blow with a hammer. Its mode of formation is varied but it usually forms in the soil profile, and under special slope, drainage and erosional conditions it may form on the surface. A laterite soil is one in which a laterite horizon is present in the profile, irrespective of its particular mode of origin, and a lateritic soil is one in the profile of which there is an incipient or immaturely developed horizon, for instance, a horizon consisting of easily recognisable ferruginous concretions and/or a mottled colouring of reddish and/or brownish iron oxides, and which, it is believed, will develop into a laterite of

favourable conditions persist long enough.' In agreement with Pendleton [1946] Preeze does not lay much stress on sesquioxide contents or the silica/alumina and silica-se. quioxides ratios. He favours a definition which does not involve chemical analysis but one which could be applied in the field.

According to Russel [1950], ferralites, the name given by Robinson [1949] to this type of soil as being more appropriate than laterities, are formed from the weathering of basic igneous rocks on well defined sites and are only deeply developed on ancient penepains which are fairly level with extensive land surfaces. They are not formed if the crust of weathering is subjected to strong ground water influences, for ground water nearly always contains water soluble silica which reacts with aluminium hydrate to form kaolinite.

A distinction has been made by Deb [1944] between laterite soils, lateritic soils and red earths. Laterite soil includes those whose profile exhibits at least one indurated slag-like or vasicular horizon, rich in sesquioxides. Lateritic soils comprise those in which no such indurated horizon is present, but which contain a considerable amount of secondary iron concretions. Red earths include the rest of the red soils which have developed under tropical and sub-tropical climates, excepting the Terra Rossa of the Mediterranean region. Laterite soils are formed under moist humid climate preferably with alternate wet and dry seasons and having water table not far from the surface. Some of the soils of this group might have formed at the tertiary lines at the level plateau with ground water table near the surface, but at present they may occupy ridges of the higher plateaus. Lateritic soils are formed under practically similar climatic conditions, but do not require alternate wet and dry seasons. The ground water level also need not be near the surface. The soil develops under more free drainage conditions. Red earths are formed under tropical and subtropical climate with a wide limit of precipitation but with perfect drainage condition. The type of soil formed depends on the amount of precipitation; low precipitation giving rise to silicate-rich soil and high precipitation giving sesquioxide-rich soil.

Deb also classifies each of these groups into three types according to the chemical composition, as allite, ferruginous and siallite with high contents of alumina, ferric oxides and silica respectively. The allite and ferruginous types are formed more easily from basic and intermediate rocks than from acidic rocks. Siallitic type, on the other hand, is formed from material preferably acidic in nature and under a comparative low rainfall.

In Bihar, the red and lateritic soils extend over a very large area covering practically the whole of Chota Nagpur. These soils, as already mentioned, are generally low in plant nutrients, yet they appear to respond well to cultivation largely because of their favourable physical properties. Careful cultural operations like manuring and drainage are, however, necessary, lest these soils, for various reasons, become poor in productivity. The purpose of the present investigation was to make a study of the properties of these soils on profile basis so as to gain information on their genesis and development and also on their classification. This included a study of

the physico-chemical properties of their clay fractions, for all soil characteristics such as permeability to air and water, their adaptability to specific crops, their fertility, absorptive power and their tilth are influenced by their colloidal constituents to a marked degree. Two typical profiles were selected for these investigations, one from the Kanke Agricultural Farm at Ranchi and the other from Hazaribagh

PROFILES

Descriptions of the two profiles are given below :

Hazaribagh soil profiles

Locality. Harenganj area of Hazaribagh town, Bihar.

Topography. Almost plain with very slight slope from West to East.

Natural vegetation. Grasses.

Cultivation. Fallow but recently brought under cultivation.

0—9 inches. Light brown sandy clay, granular in structure, slightly moist loose.

9—23 inches. Brown sandy clay, granular, with more moisture than above.

23—40 inches. As above, the colour is darker.

40—55 inches. Colour still darker, with less clayey nature.

55—72 inches. As above, very deep brown colour.

Below 19 inches dark reddish black nodules of iron were present. Grass roots were present upto 45 inches depth.

Kanke soil profile

Locality. Government Experimental Farm, Kanke, in the District of Ranchi, Bihar.

Topography. Almost plain.

Natural vegetation. Grasses as weeds.

Cultivation. Under citrus orchard.

0—8 inches. Yellow sandy clay, dry and very hard.

8—20 inches. Brown sandy loam, dry and hard.

20—32 inches. Dark brown colour, dry and hard, iron nodules present.

32—53 inches. As above, but the colour is lighter.

53—72 inches. As above, colour still lighter.

Hazaribagh has a moderate temperature throughout the year except during the hot months of April, May and June when westerly wind coming from central India brings about high temperature with very low humidity. The mean temperature rises to 85°F. in April and May, the mean maximum and minimum temperature

in these months being 99° and 76°F. respectively. In April the humidity drops to as low as 36 per cent. In winter the mean temperature is 60°F, the mean minimum being 51°F. The annual average rainfall is 53 inches, over 80 per cent of which falls during the monsoon months of June to September.

Ranchi, which is adjacent to Hazaribagh, has almost an identical climate.

RESULTS

Mechanical and chemical composition of the soils

The mechanical composition of soils from both the profiles are given in Table I.

TABLE I
Percentage mechanical composition of the soils

Profile	Depth in inches	Moisture	Loss in solution	Coarse sand	Fine sand	Silt	Clay
Hazaribagh	0—9	1.83	0.59	18.47	32.58	17.35	30.28
	9—23	2.42	0.53	13.55	26.83	18.92	37.43
	23—40	3.00	0.69	12.68	26.52	19.51	38.34
	40—55	2.65	0.61	15.60	27.20	19.45	34.31
	55—72	2.51	0.58	15.55	27.35	19.60	35.26
Kanke	0—8	1.05	0.66	33.52	36.90	11.82	17.35
	8—20	2.05	0.53	19.72	25.00	16.22	36.92
	20—32	2.53	0.67	20.39	21.74	16.11	38.74
	32—53	2.35	0.54	25.56	24.63	14.15	32.93
	53—72	1.83	0.62	30.00	19.21	17.74	32.12

In both the profiles, the clay content increases with depth, passes through a maximum value in the third layer and then decreases in the subsequent layers. This indicates that some mechanical illuviation of the soil has taken place. In the Hazaribagh profile the silt content increases slightly with depth. The same trend is followed in the Kanke profile excepting that there is an abrupt fall in the value in the 32—53 inch layer. The fine sand fraction changes irregularly with depth, but in general, it may roughly be considered to have remained constant below the first layer after an initial decrease. The coarse sand fraction at first decreases and then increases in the subsequent layers. The value of the coarse sand content of

the top layer of Kanke profile is, however, much higher and the clay content much lower than the corresponding value of the Hazaribagh profile.

Raychaudhuri and Mian [1944] who studied the red soils of Begra, Jaydebpur, Pasumalai, Midnapore and Bankura also noticed that the maximum value of the clay was in the intermediate layer, the percentage of the coarse fraction being minimum at this depth. In the case of Pasumalai profile, for example, the clay content was 56 per cent in the middle layer as compared to 35.8 per cent in the surface and four per cent in the lowest layer. The coarse sand fraction, on the other hand was only 14.5 per cent in the middle layer, the top and bottom layers containing 26.7 and 44.4 per cent respectively.

The organic carbon and total nitrogen contents of the soils are given in Table II.

TABLE II

Organic carbon, total nitrogen and C/N ratio in two profiles at different depths

Profile	Depth in inches	Organic carbon (per cent)	Total N (per cent)	C/N Ratio
Hazaribagh	0—9	0.413	0.067	6.2
	9—23	0.224	0.049	4.6
	23—40	0.107	0.049	2.2
	40—55	0.128	0.039	3.3
	55—72	0.106	0.033	3.2
Kanke	0—8	0.404	0.042	9.6
	8—20	0.321	0.046	6.9
	20—32	0.075	0.039	1.9
	32—53	0.128	0.029	4.4
	53—72	0.085	0.026	3.3

As is to be expected both the values decrease regularly with depth. The C/N ratio is low, far below the average in most cases, varying from 6.2 in the surface layer to as low as 2.2 in the third layer in the Hazaribagh profile, although the value is slightly higher in subsequent layers. Similar variation was observed in the Kanke profile.

Table III gives the chemical composition of the soils from both the profiles.

TABLE III

*Percentage chemical composition of soils
(on ignited basis)*

Profile	Depth in inches	SiO ₂	Al ₂ O ₃	Fe ₂ O ₃	CaO	MgO	K ₂ O
Hazaribagh	0—9	80.84	14.44	4.41	0.43	0.21	1.84
	9—23	75.25	16.57	5.64	0.51	0.32	2.38
	23—40	73.96	16.14	6.50	0.51	0.21	2.10
	40—55	75.71	13.90	7.68	0.43	0.21	2.29
	55—72	75.00	14.36	7.67	0.36	0.21	1.95
Kanke	0—8	80.33	10.99	2.95	0.34	0.20	3.81
	8—20	71.03	15.73	7.34	0.35	0.41	3.91
	20—32	70.69	16.39	7.76	0.35	0.31	4.06
	32—53	71.00	10.78	11.24	0.28	0.41	4.66
	53—72	71.61	15.79	5.39	0.31	0.31	4.12

In both cases the surface layer has over 80 per cent silica, which, however, decreases in the lower layers to a constant value of about 75 per cent in Hazaribagh profile and about 71 per cent in the case of Kanke. Alumina content varies from between 14 and 16 per cent and between 11 and 16 per cent in the depth, excepting that it drops abruptly in the lowest layer in Kanke. Calcium and magnesium are low. The potassium content is, however, rather high, indicating the presence of some potash bearing rocks such as micas, hornblende and feldspar.

The base exchange capacity of the soil is low and its value follows the clay content down the profile (Table IV). The exchangeable calcium which is the predominant cation increases with depth in the case of Hazaribagh soil indicating some mobilisation. In the case of Kanke it increases up to the third layer and then decreases. Exchangeable magnesium is proportionately low. Potassium is practically absent. In all these respects the soils behave in the same way as other red soils as reported by Raychaudhuri and his co-workers [1943]. The soils are slightly acidic, the pH value varying between 5.8 and 6.6. Water soluble salts are necessarily very low.

Properties of hydrogen clays

The clay fractions, less than two microns size, were prepared from the soil samples by dispersion in distilled water and siphoning off the top 20 cm. of the suspension after 24 hours. The organic matter was removed from the clay fraction by treatment

TABLE IV

Base exchange capacity and exchangeable cations in me/100 gm. soil

Profile	Depth in inches	Base exchange capacity	Exchangeable Ca	Exchangeable Mg	Exchangeable K	Water soluble salts (per cent)	pH
Hazaribagh	0-9	8.59	4.33	1.63	traces	0.06	6.13
	9-23	10.78	5.54	1.64	"	0.28	6.10
	23-40	14.40	7.14	1.65	"	0.83	5.85
	40-55	10.85	7.11	0.95	"	0.30	5.83
	55-72	10.63	7.10	0.35	"	0.20	5.94
Kanke	0-8	4.47	3.62	0.60	traces	0.05	6.21
	8-20	8.60	4.32	1.23	"	0.06	5.88
	20-32	13.74	6.18	1.44	"	0.08	5.77
	32-53	6.82	4.66	1.43	"	0.04	6.28
	53-72	7.63	4.80	0.61	"	0.03	6.63

with six per cent H_2O_2 . All exchangeable cations were then replaced by H ions by repeated leaching with 0.02 N HCl. The H clay thus obtained was washed free of excess acid and dispersed in distilled water to obtain an approximately one per cent clay in the suspension.

The chemical composition of the H clays from different layers are given in Table V.

TABLE V

Chemical composition of H clays

Profile	Depth in inches	SiO ₂	Al ₂ O ₃	Fe ₂ O ₃	CaO	MgO	K ₂ O	SiO ₂ /R ₂ O ₃	SiO ₂ /Al ₂ O ₃
Hazaribagh	0-9	50.02	33.00	16.62	traces	0.48	3.50	1.95	2.58
	23-40	49.59	36.08	12.93	"	0.53	3.26	1.90	2.34
	55-72	49.10	34.34	12.60	"	0.72	3.33	1.97	2.43
Kanke	0-8	47.30	33.55	11.16	traces	0.53	2.83	1.98	2.40
	20-32	47.48	39.00	12.04	"	0.59	3.14	1.73	2.07
	53-72	49.29	31.45	12.77	"	0.47	3.00	2.12	2.66

The values of SiO_2 , Al_2O_3 and Fe_2O_3 are in agreement with those of the clay fraction of red and lateritic soils from other localities of India as reported by previous workers [I.C.A.R. 1938]. The low values of $\text{SiO}_2/\text{Al}_2\text{O}_3$ and $\text{SiO}_2/\text{R}_2\text{O}_3$ ratios suggest that the dominant clay mineral contained in these soils is kaolinite. It may, however, be mentioned that in pure kaolinite isomorphous replacement of aluminium by iron in the crystal lattice is almost negligible [Grim, 1942] and the high content of iron found in these clays therefore shows that the greater fraction of it is probably present as free iron oxide rather than an integral part of the lattice. This, in fact, was the case, for Truog treatment [Truog, *et al.*, 1937] of the H clay from 0.8 inch layer of Kanke profile reduced its Fe_2O_3 content from 11.16 to 6.30 per cent. The magnesium content is very low and is practically negligible. Potassium is, however, rather high, being present to the extent of over three per cent K_2O , but this does not necessarily mean that the clay is illitic since both the $\text{SiO}_2/\text{R}_2\text{O}_3$ and $\text{SiO}_2/\text{Al}_2\text{O}_3$ ratios are low. If potassium did really originate from illite this mineral in the clay would have amounted to over 50 per cent according to the calculation of Grim [1942] and would therefore, have required much higher values for $\text{SiO}_2/\text{R}_2\text{O}_3$ and $\text{SiO}_2/\text{Al}_2\text{O}_3$ ratios. Moreover, magnesium content of the clays is very low and cannot account for the presence of illite. The chemical composition of these clays rather points towards their kaolinitic make-up and the potassium has probably originated from some potash bearing rocks as feldspars and micas.

Electrometric titrations of the H clays with bases were carried out according to the method of Mukherjee [Mukherjee, *et al.*, 1936]. Ten c.c. of the one per cent solution was taken in a special seven-month titration vessel, increasing amounts of the base added and the pH of the mixture measured in each case after equilibrium was attained. The arrangement was so made that the whole experiment could be carried out in an atmosphere of hydrogen. The pH was then plotted against me. base added per 100 gm. oven dried clay. Figures 1 and 2 are typical examples of titration curves of H-clays from the surface soils of Hazaribagh and Kanke respectively. In all the cases the curves for clays from different horizons have practically the same form. There is at first a comparatively sharp initial rise in pH with the addition of the base, indicating a relatively weak acid character of the soil. This initial rise is followed by flattening of the curves which show the region of buffer action. Finally there is the inflection point in the alkaline region as would be expected of an ordinary weak acid. The base exchange capacities of the H clays from different layers as calculated from the potentiometric titration curves at pH 7.0 and at inflection points are given in Table VI.

The low values of exchange capacity suggest that the principal mineral constituent in the clays is Kaolinite. This is supported by the high initial pH of the clay suspensions as also the nature of the titration curves. In the case of Kaolinite both NaOH and Ba(OH)_2 give weak acid character of the clays, as found in the present case. Montmorillonite shows weak acid character on titration with NaOH , but Ba(OH)_2 gives a strong acid character [Mitra, 1942; Mukherjee, Mitra, Bagchi, 1951]. Illite also shows weak acid character with both the bases, but the buffering is entirely different from that of Kaolinite and the nature of the titration curve is therefore

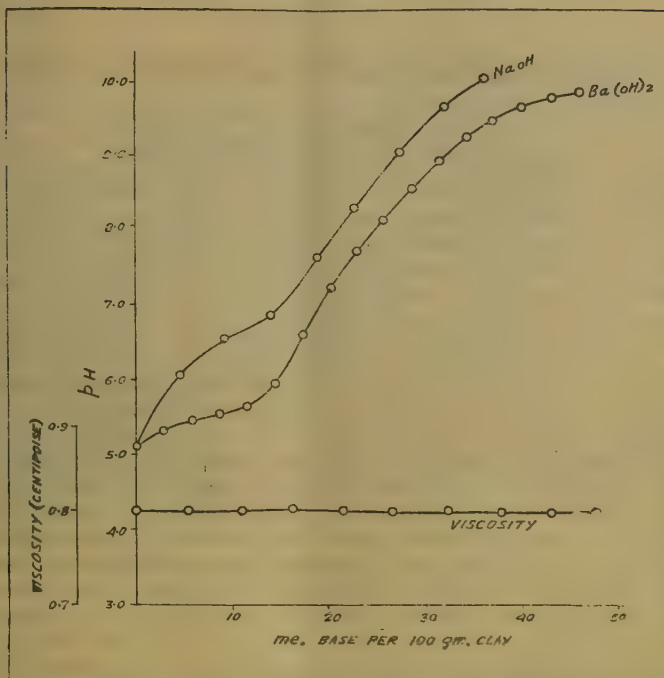


Fig. 1. Titration and viscosity curves of H-clay from 0-9 inch horizon of Hazaribagh soil

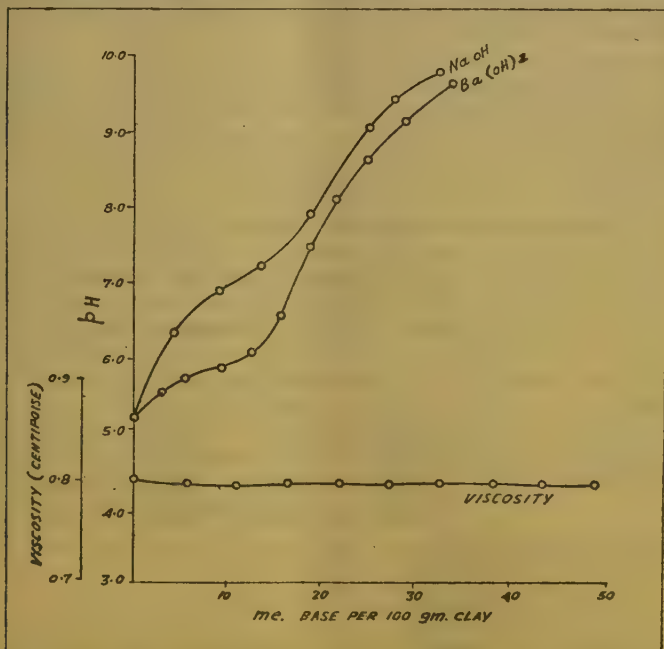


Fig. 2. Titration and viscosity curves of H-clay from 0-8 inch horizon of Kanke soil

TABLE VI
Base exchange capacity (me. per 100 gm. clay)

Profile	Depth in inches	NaOH		Ba(OH)2	
		At pH 7	At inflection	At pH 7	At inflection
Hazaribagh	0-9	12	23 (at pH 8.7)	16	17.0 (at pH 7.3)
	23-40	14	26.5 (at pH 8.4)	30	25.5 (at pH 6.3)
	55-72	11.5	25.0 (at pH 8.5)	21	23.0 (at pH 7.6)
Kanke	0-8	15	25.5 (at pH 8.7)	19.5	18.0 (at pH 6.7)
	53-72	9	19.6 (at pH 8.4)	18.0	18.0 (at pH 7.8)

different [Roy and Das, 1953]. In the present case, however, the second inflection point in the titration curve could not be obtained in any of these clays, as is requisite for a pure Kaolinite, although the titrations were continued beyond pH 10. This is because of the presence of a considerable amount of free ferric oxide in the clays, the occurrence of which masked the second inflection in the titration curves.

No change in the viscosity of the clay suspension with increasing addition of NaOH was observed (Fig. 1 and 2) which supported the conclusion arrived at previously about the Kaolinitic nature of the clay mineral, for in the case of montmorillonite, the viscosity at first increases, passes through a maximum value and then decreases with increasing base saturation [Mukherjee and Mitra, 1944]. This was confirmed by thermal dehydration studies. As in the case of pure Kaolinite, the absorbed water of the clays from soils from different layers ranged between 2.4 and 3.4, while the crystal lattice water was as high as 9.8 to 11.0 per cent.

Mineralogical composition of the sand fractions

For preparation of materials for mineralogical analysis of the sand fraction the procedure recommended by Jeffries [1941] was followed. The heavy fractions separated by bromoform. (sp. gr. 2.9) were then analysed, the results of which are given in Table VII.

TABLE VII
Mineralogical composition of the fine sand fraction of soils

Profile	Depth in inches	Heavy fraction in fine sand (per cent)	Percentage distribution in heavy fraction		
			Magnetite	Ilmenite leucoxene	Rest
Hazaribagh	0-9	2.0	53.0	6.4	40.6
	23-40	3.3	41.1	9.9	49.0
	55-72	2.9	55.7	10.7	33.6
Kauke	0-8	1.0	54.4	4.1	41.5
	20-32	0.8	50.0	9.1	40.9
	53-72	0.6	51.1	8.0	40.9

The minerals in the 'rest' group include amphibole, zircon, tourmaline, Kyanite, pyroxene, monazite, rutile and garnet in a rough order of importance. In addition to the above-mentioned minerals the soils are found to contain a considerable amount of mica, which is particularly high in the intermediate layers.

Presence of large quantities of magnetite-ilmenite, leucoxene and pyroxene, in all the horizons indicates that the parent material is a basic-basalt one. The gradual increase of leucoxene, a secondary hydrated product of magnetite-ilmenite, down the profile is expressive of a chemical eluviation to a appreciable extent.

Presence of zircon, tourmaline, kyanite, monazite and other minerals suggest a mixing of minerals of rock types in the local area, viz. Chota Nagpur, with those of the Deccan trap or Rajmahal trap which in the past occupy an area much nearer to the present locality [Auden, 1950]. Hazaribagh is located on a plateau 2000 ft. high, which is about a thousand feet lower than a pre-existing plateau to the west, remnants of which are seen in scattered elevated tracts 3000 ft. or so above the sea level. The basalts of the Deccan trap were supposed to be important rock types of this higher plateau, viz. outliers of Jashpur, Rewa and Daltanganj, from which by transportation, much material might have collected on the lower plateau of Hazaribagh and Ranchi. This may be suggested as a probable explanation for the high basalt-mineral content of these soils.

CONCLUSIONS

From the morphological features of the soils of Hazaribagh and Kanke described above, it is evident that they belong to the tropical red and lateritic soils. These soils are found under tropical and sub-tropical climatic conditions, with a wide limit of precipitation, in contrast to laterites which are formed under moist humid climate with alternate wet and dry seasons. Both the profiles show porous and friable structure and no indurated vesicular or honey combed horizon is present. Lime *kankar* and carbonates are also absent in the profiles. The drainage is free and the soluble salts are very low.

The mechanical composition of the soils agree very well with those of other similar soils reported by previous workers. The sand fraction—coarse and fine together—comprises over 50 per cent, silt forms 15-20 per cent while the rest is clay. The chemical composition shows SiO_2 to be as high as 70-80 per cent, Al_2O_3 ranges between 10-15 per cent while Fe_2O_3 which gradually increases down the profile varies roughly between 3 to 8 per cent. In consideration of the high silica content the soil may be grouped under siallite type, as suggested by Deb [1944]. Siallite type of red soil, however, usually forms from materials preferably acid in nature. The mineralogical analysis of the sand fractions of these soils, on the other hand, points towards the basic basaltic nature of the parent material. Classification of the soil into siallite group must, therefore, be made with some reservation. The parent material also contains high potash bearing minerals which accounts for 2-4 per cent K_2O in the soils. The base exchange capacity of the soil is low and calcium is the predominant exchangeable cation accounting for 44 to 80 per cent of the total exchange capacity while magnesium forms 3 to 20 per cent.

The chemical composition, electro-chemical properties, electroviscous and thermal dehydration studies all taken together show that the clay mineral constituent of the red and lateritic soils of Bihar is predominantly kaolinite. The mineral composition is practically uniform throughout the profile and no appreciable variation in depth was observed. A considerable amount of free sesquioxides, is, however, present in the clay fraction. The climatic condition under which the soils have formed by weathering, and also the nature of the parent rocks are suited to the development of Kaolinitic type of clay mineral, and in the present studies, this in fact, has been found to be the case.

The physical and chemical properties of soils ultimately depend upon the nature and amount of clay minerals contained in them and it is natural that for classification of soils the basis should be the mineralogical make-up of the clays. In the present investigation some correlation between the properties of red and lateritic type of soils of Bihar and their clay mineral constituent has been made. In India soil survey has so far been based on fertility status and the physical properties of the soils, and such correlation, therefore, points out the importance of mineralogical analysis of the clay fraction for permanent soil classification.

SUMMARY

Studies of two red and lateritic soil profiles, one from Hazaribagh and the other from Ranchi (South Bihar) have been reported. The soils were examined for mechanical and chemical compositions, organic carbon, total nitrogen, reaction, soluble salts, base exchange capacity and individual bases. Hydrogen clays were prepared from horizon samples of the profiles and their chemical composition, electrochemical, electroviscous and thermal dehydration properties showed that the dominant clay mineral in them is Kaolinite. There was practically no variation in the mineral constituent down the profile.

ACKNOWLEDGMENT

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UTILIZATION OF JUNGLE DRIED BONES

By P. C. JUNEJA, K. C. GULATI and S. P. RAYCHAUDHURI, Indian Agricultural Research Institute, New Delhi

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LARGE quantities of bones become available in India annually from animals which die of natural causes. Major part of these raw bones is exported for certain considerations, one of the considerations being lack of proper utilisation within the country. The main difficulty in the utilisation of this rich phosphatic and nitrogenous fertilizer is the inability of the cultivators to get a finely crushed product at a suitable price.

Two simple processes by which bones can be rendered readily friable are either treatment with steam or with chemicals. In the treatment of bones with steam, it is very desirable that the glue produced as a by-product be of marketable quality to establish the process on sound economic footing. But in the treatment of bones in a simple type of externally heated autoclave, it has been experienced that it is very difficult to exercise temperature control, so essential at different stages of glue extraction, although, it is well-known that better temperature controls give better quality glues. Taking into consideration various technical and economic difficulties involved, the process is of doubtful advantage as a cottage industry.

There are two chief types of chemicals, *viz.* alkalies and acids, which can be used for reaction with bones. Among the alkalies, caustic soda is outstanding and is also indigenous and in the acid type chemicals, hydrochloric acid is more indigenous than sulphuric acid.

In the treatment of bones with caustic soda, the nitrogenous part of the bones is dissolved out by the alkalies and the residual bones are rendered fragile. The process suffers from the disadvantage that the nitrogen-bearing constituents dissolved out by the alkali are lost in an unrecoverable form and for any satisfactory solution of the problem, this nitrogenous portion which is one of the major constituents of bones should be properly accounted for.

Treatment with hydrochloric acid has many advantages over other processes. Synthetic hydrochloric acid is manufactured in the country at places where the electrolytic caustic soda industry already exists. There is low production of the acid due to its moderate demand and also the production of caustic soda is limited due to the limited off-take of chlorine. As a matter of fact, lack of economic outlet for chlorine is the real bottle-neck in the development of caustic alkali industry in the country. From private communications it has been learnt, that the cost of production of 33 per cent hydrochloric acid will not exceed rupees three to four per ton at the site.

Treatment of bones with hydrochloric acid excels other processes due to the simplicity of the operation. At room temperature, one pound of 22°Be hydrochloric acid diluted to five per cent will suffice for every one pound of jungle dried

bones of under one inch size. The treatment can be carried out in wooden vats for a period of one week with occasional agitation. Under the conditions, hydrochloric acid acts upon the phosphatic part of the bones. The extracted acid phosphate and phosphoric acid are then converted to dicalcium phosphate—a well-known phosphatic fertilizer by addition of requisite quantity of milk of lime. The precipitated dicalcium phosphate after filtration is dried in the sun; the yield being 45 per cent on the weight of bones.

The residual bones obtained after the acid treatment are washed with water and subsequently dried; the yield being 40 per cent of the original weight. These are very readily friable, contain 8 per cent nitrogen and 20 per cent P_2O_5 of which two-third is citrate soluble. This is more or less a balanced fertilizer (bone manure). Sixty per cent of the nitrogen of the manure is nitrifiable after eight weeks, thus making it a good nitrogenous fertilizer.

Thus a simple process has been evolved for the utilization of bones to yield dicalcium phosphate and a balanced bone manure, and has scope of being used on a small industrial scale.

RATOONING OF SUGARCANE AS INFLUENCED BY THE TIME OF HARVESTING PLANT CANE

By R. K. TANDON, G. P. KAPOOR and G. N. MISRA, Main Sugarcane Research Station, Shahjapur, U. P.

(Received for publication on 19 November 1953)

(With Plate I)

TILL recently there had been a considerable controversy on the problem of ratooning of sugarcane and the suitability of the practice under North Indian conditions. The investigations carried out under the Ratooning Scheme at Kalai (District Aligarh, U. P.)* have, however, solved the problem to some extent. It has been established that ratooning limited upto first year ratoon only, is a justifiable practice and can usefully be adopted by the cane growers. Under proper care and management, a ratoon crop is capable of giving as high yields as plant cane and the practice does not necessarily lead to an increase in the incidence of pests and diseases. It has also been confirmed that the ratoon crops mature early and are capable of giving better recovery in the early part of the season.

Following these results, it became pertinent to evolve proper manurial, cultural and irrigational schedules for the ratoon crop, as it is generally accepted that the latter behaves unlike the original crop of plant cane, under varying soil and climatic conditions. The manurial requirement of a ratoon crop has already been determined [Kapoor, 1952] and the present paper is confined in its scope to the study of cultural aspect only.

PRELIMINARY OBSERVATIONS

When a plant crop is harvested, the stubbles along with a network of old root system are left in the field. Tapering towards the base, these lower-most portions of cane stems are composed of short joints, each provided with a bud more or less in a dormant condition. After the harvesting of the crop when the conditions become favourable, some of these buds strike out forming into primary shoots and later developing a number of secondary and tertiary ones. Each of these shoots develops a root system of its own emerging out from the lower-most root ring.

The germinating buds in a ratoon crop are situated very near the ground surface and are placed in almost a vertical position, whereas in the plant crop they are deeper in the soil and placed in a horizontal position. The set-roots and the shoot-roots in the ratoon crops have thus to cover a considerable distance before reaching the soil moisture level. The older root system eventually dies out and the new root system of the ratoon crop remains at a shallower depth as compared to that of

* Final report of the work done under the Sugarcane Ratooning Scheme at Kalai (Aligarh)—1939-49; Superintendent, Printing and Stationery, Allahabad, Uttar Pradesh, India.

the plant crop (Plate I, Fig. 1). The tillers develop at a high level, more so in cases where the canes are harvested without dismantling of ridges, thus leaving a greater length of the original stems underground (Plate I, Fig. 2). It is generally observed that the top-most bud which is nearest to the cut end of the stem is often the first to strike out.

The crushing season in Uttar Pradesh usually commences about the beginning of December and continues upto April or even later upto May. During this period, there is a change over of the weather conditions from one extreme to the other; December and January are the coldest months of the year, February and March moderately cold and the period from April to June is extremely hot (Appendix I). Growth and development in a crop are dependent on a number of external and internal factors. The time of harvesting of plant cane which virtually marks the beginning of ratoon may, therefore, having a special bearing on the performance of the ratoon. It may be so particularly because harvesting season in sugarcane normally extends over a very long period when there is marked variation in the weather conditions. Dutt [1950] stated that the parent crop harvested early in the season was believed to give poor sprouting specially in frosty areas and emphasized the necessity of finding out the optimum time of harvesting the plant cane to secure the maximum advantage from ratoons.

EXPERIMENTAL

The experiment was conducted on first year ratoon crops of the popular cane varieties, Co. 313, Co. 421 and Co. 453 during the three consecutive crop seasons, 1949-50, 1950-51 and 1951-52. Variety Co. 421 was, however, replaced by another popular variety, Co. S. 186, in the last two seasons as the former was noticed to be deteriorating under normal standard of cultivation. Originally it was planned to harvest the crop at monthly intervals extending from mid-December to mid-April but owing to the closing down of the neighbouring sugar factory earlier than April, the plots reserved for harvesting during the latter month had to be harvested in the month of March in the first two seasons. In the third season, however, the crop under all the treatments could be harvested at the scheduled timings. The treatments were replicated five or six times in a split-plot design with main plots to dates of harvesting plant cane and sub-plots to varieties.

Observations with regard to the formation of shoots, millable canes available at harvest, juice quality of the crop and the final yield per acre under each treatment were recorded every year. The data collected did not show any interaction between the varieties and the dates of harvesting during any of the three seasons. Discussions in this paper are, therefore, confined only to the results obtained with different dates of harvesting plant cane, irrespective of the varieties tested.

RESULTS

Shoot production. In order to obtain a comparative estimate of the number of shoots produced per acre under different treatments, shoot countings recorded in June during all the three crop seasons are given in Table I.

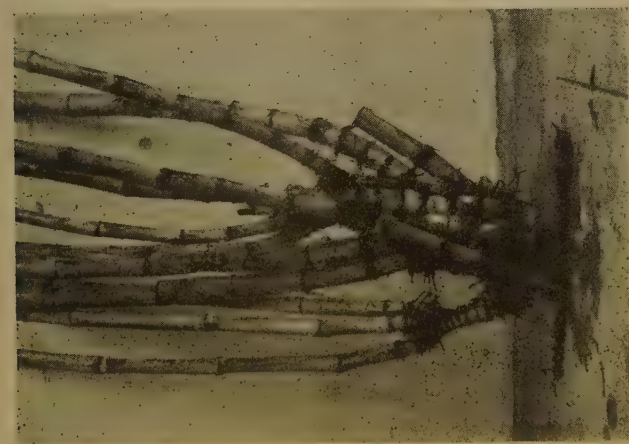


Fig. 1. Older root system dies out and the new system of the ratoon remains at a shallower depth

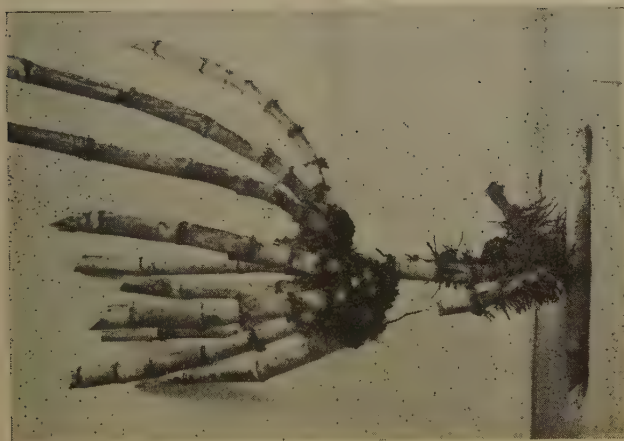


Fig. 2. Tillers develop at a high level leaving a greater portion of the original stem underground

TABLE II

MONTH AND DATE OF HARVESTING PLANT CANE	DATES OF SHOOT COUNTINGS						
	29-1-51	28-2-51	31-3-51	1-5-51	1-6-51	2-7-51	28-7-51
DECEMBER ^E 17-12-50	43 _x 330 _{xx} 14197 _{xxx}	73 271 22323	105 552 39981	136 (694) 61510	167 (598) 80051	198 189 85917	224 -662 68699
JANUARY 16-1-51		43 712 30624	75 550 48224	106 592 66587	137 626 86005	168 135 90200	194 -600 74595
FEBRUARY 15-2-51			45 1173 52800	76 671 73597	107 (660) 94072	138 -70 87912	164 -609 72072
MARCH 9-3-51				53 1475 78173	84 (686) 99440	115 -38 98267	141 -732 79229
APRIL 22-4-51					40 (2617) 104661	71 372 116189	97 -945 91608

Absolute rate of production and accumulated number of shoots per acre on different dates in relation to the age of the crop

X—Number of days after harvesting plant cane, indicating the age of the ratoon crop.

XX—Number of shoots produced per acre per day during the interval preceding the date of shoot counting.

XXX—Number of shoots accumulated per acre.

NOTE—Vertical columns show a comparison of the date as observed on different dates while oblique columns are meant to compare the results for ratoons of approximately the same age.

TABLE I

Comparative number of shoots in thousands per acre

Crop season	Treatments (Time of harvesting plant cane)					S. E./acre
	Mid-December	Mid-January	Mid-February	Mid-March	Mid-April	
1949-50	55.71	80.10	73.64	70.93	..	3.26
1950-51	64.79	73.32	78.70	82.89	..	2.21
1951-52	80.05	86.01	94.07	99.44	104.66	5.06
Mean	66.85	79.81	82.14	84.42	104.66	..

It is thus evident that the number of tillers produced per acre by the ratoon crops increased in general with the delay in harvesting of plant cane. Harvesting in December invariably resulted in the production of lowest number of shoots per acre as against the treatments, where harvesting was done at later dates. Maximum number of shoots were, however, recorded in mid-March treatment during the second year and in mid-April treatment during the third year of experimentation. The differences of tiller counts between February and March harvests during the years 1950-51 and 1951-52, wherein the varieties remained unchanged, are not statistically significant.

It was also observed that the rate of production of shoots under different dates of harvesting varied considerably. Detailed observations in this respect were recorded during the year 1951-52 and are presented in Table II.

Progressive shoot counts show that in the December and January started ratoons after the first flush of shoots, there is somewhat a fall in the rate of shoot production per day which is followed by a gradual rise till about the month of May. This is, however, not observed in the ratoons started later.

Comparing ratoons of approximately equal ages, through oblique columns of Table II, it is seen that the number of shoots produced per day per acre was progressively higher to start with as the date of harvesting plant cane was delayed from December to April, 330, 712, 1173 and so on. The superiority of comparatively later dates of harvesting in the number of shoots produced per day was most in evidence at an age of about six weeks where it was observed to be 2617 against a

much smaller number in the earlier harvested ratoons. Another more important fact is discernible from Table II; irrespective of when the ratoon is started, all the way from December to April, the maximum number of shoots formed per day occurring in the ratoons of approximately equal ages (oblique column) are observed only in the month of May (shown under 1-6-51) as shown by the figures in circle.

In addition to the study on the progress of shoot production, the number of clumps present at harvest of the plant cane and those which actually formed the following ratoon crop were counted during the crop season 1951-52. The percentage mortality of clumps was estimated for each treatment on the basis of the above data. The results obtained are given in Table III.

TABLE III

Percentage mortality of clumps in the ratoon crops under different treatments

Treatments (Time of harvesting plant cane)				
Mid-December	Mid-January	Mid-February	Mid-March	Mid-April
6.23	4.95	1.92	0.72	0.44

December and January harvested crops resulted in the heaviest mortality of clumps as compared to the crops harvested at later dates. As the harvesting was delayed, the percentage mortality was gradually reduced so much so that less than one per cent mortality was recorded in crops harvested during the months of March and April. Increased space available in between the clumps, generally results in inducing heavier tillering but the loss due to heavy mortality in the crops harvested during December and January was not compensated, as the average number of tillers per clump under these treatments remained lower than those where the crops were harvested at later dates (Table IV).

TABLE IV

Average number of shoots per clump during the month of June, 1951

Treatments (Time of harvesting plant cane)				
Mid-December	Mid-January	Mid-February	Mid-March	Mid-April
11.2	11.4	12.2	12.2	13.6

Millable canes. The number of shoots produced per acre was at its maximum by about the break of rains after which, during the rainy season the population per acre was reduced to nearly fifty per cent. Mortality of shoots was comparatively higher in the plots where the plant crop was harvested at later dates than in plots wherein the crop was removed earlier. Total number of millable canes per acre formed under different treatments are given in Table V.

TABLE V

Average number of millable canes in thousands per acre under different treatments

Crop Season	Treatments (Time of harvesting plant cane)					
	Mid-December	Mid-January	Mid-February	Mid-March	Mid-April	S.E. per acre
1950-51	37.35	41.98	42.36	41.57	..	0.931
1951-52	38.75	38.98	42.09	43.44	43.00	..
Mean	38.25	40.48	42.23	42.51	43.00	..

Statistical analysis of the data revealed that the December harvesting treatment produced the lowest number of millable canes per acre in comparison with other treatments which were rather equal among themselves ; the slight differences amongst them being statistically insignificant. During 1951-52, however, even the December harvesting failed to exhibit any substantial difference, although the number of millable canes per acre was again the lowest in this season as well.

Yield per acre. Average yield of cane per acre under the different treatments is given in Table VI.

TABLE VI

Average yield of cane per acre under different treatments in maunds

Crop Season	Treatments (Time of harvesting plant cane)					
	Mid-December	Mid-January	Mid-February	Mid-March	Mid-April	S.E. per acre
1949-50	533.6	563.6	617.6	605.1	..	17.52
1950-51	344.4	419.7	427.6	441.8	..	18.97
1951-52	424.2	449.8	472.7	503.2	443.2	22.51
Mean	434.1	477.7	506.0	516.7	443.2	..

Ratoons developed after harvesting of plant cane during December have consistently given the lowest yields of cane in comparison with those started at later dates. Although the tillering in ratoon crops improved progressively with the delay in the harvesting of the plant cane from December onwards up to April, the highest cane yields were obtained, only by taking ratoons from February or March harvested crops of plant cane, April treatment giving only as good yields of cane as January treatment. This is corroborated by the data presented in Table VII which show that the April treatment had canes of shorter length and poorer weight in comparison with January, February and March treatments.

TABLE VII
Average length and weight of canes at harvest 1951-52

Particulars	Treatments (Time of harvesting plant cane)				
	Mid-December	Mid-January	Mid-February	Mid-March	Mid-April
Length per cane in inches.	58.5	62.4	62.2	62.9	58.3
Weight per cane in chattaks.	7.0	7.4	7.2	7.4	6.6

Juice quality. The average juice quality data for each treatment are given in Table VIII.

TABLE VIII
Seasonal average juice quality of canes

Crop Season	Treatments (Time of harvesting plant cane)				
	Mid-December	Mid-January	Mid-February	Mid-March	Mid-April
<i>(a) Sucrose per cent juice</i>					
1949-50	13.61	14.10	13.97	14.32	..
1950-51	15.39	15.66	15.69	15.69	..
1951-52	15.01	15.18	15.38	15.14	15.60
Means	14.67	14.98	15.01	15.05	15.60
<i>(b) Purity coefficient</i>					
1949-50	79.34	82.51	81.64	81.47	..
1950-51	83.81	85.70	85.88	85.40	..
1951-52	81.22	82.52	83.53	82.64	84.25
Means	81.46	83.58	83.68	83.17	84.25

The results consistently indicate a somewhat inferior juice quality associated with the December started ratoons in comparison with those started later. This effect is particularly marked in the figures for the purity coefficient.

DISCUSSION

It is clear from the data presented in the foregoing pages that the temperatures of the soils as obtained in the central parts of Uttar Pradesh during the months of December and January act as a limiting factor resulting in very slow progress of the production of shoots. While it has been pointed out that sprouting of buds takes place to some extent even at as low a temperature as 43°F [Sartoris, 1929] ; Dillewijn [1952] after quoting the findings of some more workers, Verret [1927], Rege and Wagle [1939], Ryker and Edgerton [1931], inferred that germination is very slow at temperatures below 70°F and improves progressively up to a temperature of 80 to 90°F. Further corroboration on this point is received from the work of Mathur [1941] who found that the critical temperature for germination was nearly 66°F, below which germination was inhibited. He further observed that the temperature of 92 to 93°F attained in central U. P. during May seems to be favourable for tillering in sugarcane.

As the date of harvesting plant cane was delayed, the ratoons getting more favourable temperature conditions from the very start, produced tillers comparatively at a faster rate. The early started ratoons having passed through a stage of unfavourable low temperature conditions could not produce tillers as many in number as that in later started ratoons, even when favourable temperature conditions became available. It thus appears that low temperature at the early start does not only delay the production of shoots, but also inhibits the sprouting of buds permanently by adversely affecting the stubbles. There is, therefore, higher mortality of clumps under winter started ratoons than those started later. It also stands to reason, since sugarcane is a tropical plant prone to thrive better under warmer rather than cooler conditions. The greater slowing down of the production of shoots in the comparatively later started ratoons during June, is apparently due to closing up of the rows caused by profuse tillering at a time, when early started ratoons were more open. This higher rate of production of tillers in early started ratoons, during the late season could not continue for long, due to close of the tillering phase imposed by the onset of rainy season when the conditions were more favourable for the elongation of stalk. Thus the initial setback in the tillering phase appears to have ultimately an adverse effect on the crop.

With the onset of rainy season, elongation of stalk and leaf expansion takes place at a rapid rate. This results in a greater struggle for existence, causing heavy mortality of shoots in almost all the ratoon crops in general and later started ratoons in particular. The existing differences between the treatments in the number of millable canes per acre were thus masked to a considerable extent. But the December started ratoons still remained markedly inferior to the later started ones both in respect of the millable canes and the final yield per acre.

RECOMMENDATIONS

For securing optimum results in respect of both yield and sugar per acre from the first year ratoon crop, plant cane should be harvested towards or after the close of the winter season. If harvested earlier, the development and growth of ratoon crop is adversely effected, resulting in poorer juice quality and lower yields of cane. Undue delay, *i.e.* after the month of March or at the most April, in harvesting plant cane is also undesirable, as it is necessary that the plant crop should be harvested well ahead of the onset of rainy season. It is only then that the full advantage of the favourable weather conditions for the growth of the crop during rainy season can be derived. The months of February and March are thus the most favourable time for harvesting plant cane from the fields where it is intended to keep ratoons.

SUMMARY

An experiment was conducted at the Main Sugarcane Research Station, Shahjahanpur, during the three crop seasons from 1949-50 to 1951-52, to ascertain the proper time of harvesting plant cane for securing maximum yield of ratoon crop. The treatments consisted of five dates of harvesting plant cane of three popular varieties at monthly intervals from mid-December to mid-April.

There appeared no interaction between the different varieties of cane and the dates of harvesting cane.

The rate of tillering in ratoon crops increased with the delay in harvesting from December to April. In cases where plant crops were harvested during December and January, heavier mortality of cane stumps was noticed as compared to the treatments in which the ratoon crops started at later dates.

There was heavy mortality of tillers during rainy season in all the ratoons but it was more conspicuous in those which started later, with the result that the initial differences in the number of tillers per acre between different treatments were greatly minimised. December started ratoons, however, showed the worst performance and the number of millable canes at harvest were the lowest under this treatment as compared with others.

Contrary to what is found in plant cane when the growth period of the crop is lengthened, December started ratoons indicated a somewhat inferior juice quality (sucrose and purity) than ratoons started from January to April.

Best cane yields were obtained from ratoon crop taken after harvesting plant cane in February or March, while December started ratoon gave the lowest yields of cane per acre.

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APPENDIX I

Meteorological data recorded at the Main Sugarcane Research Station, Shajahanpur

Year	January	February	March	April	May	June	July	August	September	October	November	December
<i>Mean minimum temperature in °F.</i>												
1949	45.6	50.5	58.0	60.2	78.6	81.5	79.4	78.3	77.1	65.2	49.0	42.8
1950	47.2	46.9	56.2	65.9	76.2	79.0	78.6	77.4	74.4	61.4	48.9	48.8
1951	48.1	48.5	57.6	65.7	76.3	80.0	80.2	78.8	75.4	60.3	53.6	48.0
<i>Mean maximum temperature in °F.</i>												
1949	75.4	77.0	88.9	99.9	105.0	105.8	92.3	88.9	89.8	89.8	79.8	78.1
1950	72.6	74.4	85.8	97.7	104.7	99.9	91.8	90.1	90.2	92.1	82.3	73.4
1951	70.2	77.6	88.3	96.1	104.1	102.9	97.7	92.6	91.3	96.2	85.4	78.1
<i>Mean temperature in °F.</i>												
1949	58.7	62.8	72.1	52.3
1950	57.1	57.5	71.6	86.4	97.0	94.5	88.6	85.8	88.8	73.1	59.1	49.4
1951	48.0	55.7	70.3	80.9	92.3	98.2	89.5	84.8	88.4	77.6	61.6	53.5
<i>Rainfall in inches</i>												
1949	..	0.92	0.02	0.16	0.01	0.08	14.58	16.43	15.00	1.46
1950	1.20	0.25	0.23	0.01	0.38	4.43	10.89	12.60	6.79	0.72
1951	1.96	0.64	1.15	0.07	0.10	2.84	1.61	10.12	12.20	0.03	0.74	..

CYTOGENETICAL INVESTIGATIONS ON GROUNDNUTS

I. THE SOMATIC CHROMOSOMES*

By C. N. BABU, Sugarcane Breeding Institute, Coimbatore

(Received for publication on 31 December 1953)

(With one text-figure)

GROUNDNUT, *Arachis hypogaea* L., is cultivated in many countries of the world, mainly for its pods which are of great utility to the common man and the industrialist alike. The original home of the plant is supposed to be Brazil, South America, where its allied species are known to grow in the wild state, while *A. hypogaea* itself has not been found growing wild anywhere. About a dozen species were described by earlier systematists. Of these, only *A. hypogaea* has gained prominence due to its world-wide commercial importance. There are innumerable forms and varieties of cultivated groundnuts which differ among themselves in a number of characters, vegetative, floral, fruiting and economic, so much so that the species *A. hypogaea* itself has been subject to taxonomic and cytological analysis. A survey of the vast number of its forms and varieties has been considered almost tantamount to a survey of the genus itself. For, from such a survey have emerged discussions regarding not only the origin and classification of groundnuts, but also in particular about the taxonomic position of certain varieties like *A. nambyquaroe* and *A. rasteiro* of doubtful specific status.

CYTOLOGICAL LITERATURE

The chromosome numbers of many forms of *A. hypogaea*, some of its allied species and several artificial hybrids, have been recorded by several authors [Badami 1928, Kawakami 1930, Husted 1933, 36 and Mendes 1947]. The numbers determined by these authors indicate polyploidy in the speciation of *Arachis*, with both $2n=20$ and $2n=40$ occurring among them. Husted [1936] studied the somatic chromosome morphology and meiotic behaviour and inferred that the runner and bunch varieties must have had a common origin and that *A. nambyquaroe* and *A. rasteiro* must be designated as only sub-species of *A. hypogaea*. Mendes [1947], observing a chromosomal chimera with 80 somatic chromosomes in a 40-chromosome form, considered it as an indication of the tetraploid nature of the latter, having been derived by doubling of the chromosomes in the diploid species with $2n=20$. Further, the occurrence of chromatin bridges, lateral trivalent, chromosome ring and fragments in somatic mitoses of certain groundnuts, as also somatic doubling of chromosomes which remained in paired condition after doubling were recorded (Babu 1941), the former phenomena indicative of structural changes and the last mentioned feature lending support to Mendes's observation.

*This paper forms part of the work done by the author in 1940, as a Research Fellow of the Madras University. The author's thanks are due to the Madras University for the award of a stipendiary fellowship

MATERIAL AND METHODS

The Groundnut Breeding Station at Tindivanam maintains a good world-wide collection of cultivated groundnut varieties and forms under the control of the Oil Seeds Specialist, Coimbatore, and the material for this study was obtained from that centre. A preliminary examination of a few types revealed some striking differences in the somatic chromosome morphology, whereas Husted [1936] had observed a general similarity between the chromosomes of the types he studied. Hence a critical survey of the somatic chromosomes of all the available forms of groundnuts from the point of view of chromosome morphology was planned. At the outset, however, the examination was restricted to certain selected types, possessing a fair representation of the variations in the external morphological and economic characters. In making such a choice, the 'Classification of Groundnut varieties' by John, Narayana and Seshadri (unpublished) was referred to. Particulars of the types so selected are given below:

No.	Type	Habit	Nomenclature
1	Kumavu	Bunch	<i>A. hypogaea</i> var. <i>oleifera</i> (valentia s. gr.)
2	Florida-V	Bunch	<i>A. hypogaea</i> var. <i>oleifera</i>
3	Form A. H. erecta	Erect	<i>A. hypogaea</i> var. <i>oleifera</i> forma <i>erecta</i>
4	Spanish Philippine	Bunch	<i>A. hypogaea</i> var. <i>oleifera</i> (Java s. gr.)
5	Kurumani	Semi-spreading	<i>A. hypogaea</i> var. <i>asiatica</i> (asiatica s. g.)
6	H. G. 1	Semi-spreading	<i>A. hypogaea</i> var. <i>oleifera</i> (Native Tanganyika sub group)
7	Tennessee Red	Bunch	<i>A. hypogaea</i> var. <i>oleifera</i> (valentia)
8	Gudiathum Bunch	Bunch	<i>A. hypogaea</i> var. <i>oleifera</i> (Spanish)
9	Madagascar	Spreading	<i>A. hypogaea</i> var. <i>oleifera</i> (Big Japan)
10	Var. gigantea	Trailing	<i>A. hypogaea</i> var. <i>gigantea</i> (trailing)
11	Local Mauritius	Spreading	<i>A. hypogaea</i> var. <i>oleifera</i> (L. Mauritius)
12	'A. nambyquaroe'	Semi-spreading	<i>A. hypogaea</i> var. <i>nambyquaroe</i> (nambyquaroe-Rasteiro s. g.)
13	'A. rasteiro'	Spreading	<i>A. hypogaea</i> var. <i>nambyquaroe</i> (nambyquaroe-Rasteiro s. g.)
14	Corientes-3	Bunch	<i>A. hypogaea</i> var. <i>oleifera</i> (Corientes 3)
15	F ₁ of (Corientes-3 X Local Mauritius)	Spreading	Hybrid

Root tips from germinating pods were fixed in Levitsky's fluid (six parts of 10 per cent formalin and five of 1 per cent chromic acid). Sections were stained by the Feulgen fast green technique. As comparison was intended, strict uniformity

was observed in every procedural detail right from the time of fixation of the root tips up to the completion of the camera lucida drawings. The latter were made at table level obtaining a magnification of 2900, suitably reduced in print.

OBSERVATIONS

The 15 plants examined have 40 somatic chromosomes each. The majority of the chromosomes have median constrictions and exhibit a range of size. In these, as well as in the possession of a pair of conspicuously small chromosomes known as the 'A' pair [Husted 1936], (Fig. 1), the types examined exhibit similarity. This is in keeping with Husted's observations. But the 'B' chromosomes (Fig. 1), with unusually long secondary constriction noticed by Husted in all the types he examined, are present in only half a dozen forms of this investigation (Fig. 6 to 9 and 12 and 13). In some of the forms a pair of chromosomes with only a slightly long secondary constriction is recognisable (Fig. 6 and 10 to 13). This is designated the B_1 chromosome in the present study (Fig. 1). Another type of secondary constricted chromosomes, S, is present in some complements (Fig. 8, 12 and 16). This S chromosome would appear to be different from the B_1 chromosome mainly in respect of the secondary constriction. Barring Tennessee Red, the types possess a pair of satellited chromosomes each (SAT). These particulars as well as the number of telophasic nucleoli formed in each case are given below:

Type	Somatic complex	No. of telophasic nucleoli
1. Kumavu	2Sm, 2SAT, 34M, 2'A'	2
2. Florid V	2Sm, 2SAT, 34M, 2'A'	4
3. Spanish Philippine	2Sm, 2SAT, 34M, 2'A'	2
4. Kurumani	2Sm, 2SAT, 34M, 2'A'	4
5. H.G. 1	2Sm, 2SAT, 34M, 2'A'	4
6. A. H. erecta	2S, 2SAT, 34M, 2'A'	4
7. Tennessee Red	2'B', 2Sm, 34M, 2'A'	4
8. Gudiathum Bunch	2'B', 2SAT, 34M, 2'A'	4
9. Madagascar	2B ₁ , 2SAT, 34M, 2'A'	4
10. A. H. Gigantea	2B ₁ , 2SAT, 34M, 2'A'	4
11. Local Mauritius	2'B', 2B ₁ , 2SAT, 32M, 2'A'	6
12. 'A. nambyquaroe'	2'B', 2B ₁ , 2SAT, 32M, 2'A'	6
13. 'A. rasteiro'	2'B', 2S, 2SAT, 32M, 2'A'	6
14. Corientes-3	2B ₁ , 2'B', 2S, 2SAT, 30M, 2'A'	8



Fig. 1 to 16:—Figures have been drawn at a magnification of $\times 2900$ except fig. 1 which is diagrammatic representation of the types of the chromosomes noticed in groundnut. Although the constrictions of the chromosomes are actually transparent, they have not been left blank in the figures in order to prevent confusion in following the individuality of the chromosomes.

Fig. 2 to 16:—Somatic complements; (Fig. 2. Kamavu; 3. Florida V; 4. Kurumani; 5. Spanish Philippine; 6. *A. nambyquaroe*; 7. Tennessee Red; 8. *A. rasteiro*; 9. Gudiathum Bunch; 10. Madagascar; 11. *A. h. Var. gigantea*; 12. Corientes; 13. Local Mauritius; 14. F1 of a cross between Corientes 3 and Local Mauritius; 15. H. G. 1; 16. *A. h. forma erecta*).

From the above it may be seen that in most cases correspondence exists between the number of telophasic nucleoli and the number of 'B', B₁, S and/or SAT chromosomes present in each case. The association of these chromosomes with the prophase nucleolus was studied in detail but the results have been intended for a separate self-contained article on the subject. It is only desired to point out here that while the groundnut types exhibit general similarity in respect of the 'A' and M chromosomes, the nucleolar ones, viz. the 'B', B₁, S and SAT chromosomes are not universal in their presence in the groundnut types.

Yet another type of differentiation is noticeable among the somatic complements of the different varieties. This is in respect of the overall size of the chromosomes of the entire complement. Hunter and Leake [1930] reported that Badami [1928] had found 'Small Japan', a bunch type, to possess almost double the size of chromosomes in the procumbent type, 'Virginia.' Similar, if not to such a great extent, size difference is perceptible between spreading and bunch types having more or less the same chromosome make up in the somatic complement. For instance, the chromosomes of 'Kurumani' and 'H.G.1,' both semi-spreading types (fig. 4 and 15), are on the whole, of significantly smaller size, particularly in length, than those of either 'Kumavu,' 'Florida-V' or 'Spanish Philippine' (bunch). All these have more or less the same chromosome make up, though there may be some slight differences in the median constricted chromosome sets. Further, it may be seen from the figures that 'A. H. Gigantea', a trailing form evolved from a cross between 'Gudiathum Bunch' (bunch) and 'Madagascar' (spreading) has comparatively shorter chromosomes than those of 'Gudiathum Bunch.' (The chromosomes of 'Madagascar' may be seen to be much thinner than those of 'Gudiathum Bunch'.) Similarly, the chromosomes of the F₁ plant (spreading) of a cross between 'Corientes-3' (bunch) and 'Local Mauritius' (spreading) are much shorter than those of the bunch parent. And finally, the evolved type forma erecta, with an erect habit possesses much bigger chromosomes than those of 'H.G.1' (semi-spreading), one of the parents of the cross from which it was evolved, the other parent being a bunch type.

DISCUSSION

Varietal classification

Van der Stock [1910], Chevalier [1933] and Hays [1933] have attempted systematic classification of groundnut varieties using characters like duration, growth habit, branching, pods, kernels, etc. Luzina [1935], after examining nearly 300 specimens, collected from various parts of the world, expressed insufficiency of both the material as well as biological information to facilitate a clear classification. However, John and others have succeeded in classifying groundnuts, into seven main groups comprising five varieties and 23 forms, after carrying out a thorough systematic and genetic survey of the collections maintained at the Tindivanam Groundnut Breeding Station. In the main grouping, growth habit has been utilised, while testa colour, size and nature of pods, and kernels have been used as further classificatory factors. Cytological findings have been hitherto applied to for inferring the probable mode of origin of certain forms of the groundnut, as also for deciding on the position of some types like *Arachis nambyquaroe*. It would be clear from the present study that a

closer understanding of the genus *Arachis* itself from classificatory and genetical point of view may be made possible by a thorough examination of the somatic chromosome morphology of all the available varieties and forms of groundnut. The correlation noticeable between genotypic change and the growth habit is particularly interesting in view of the fact that the growth habit has been utilised as a major classificatory factor by John and others.

SUMMARY AND CONCLUSIONS

1. A critical survey of the somatic chromosome morphology of 14 varieties and forms of groundnut has been made.

2. There is a general similarity between these types in respect of the median constricted chromosomes which form the majority in the somatic complement.

3. A pair of long chromosomes with unusually long secondary constrictions is recognizable in some types, while in the others, ordinary secondary constricted chromosomes may be present. Satellited chromosomes are present in all the types except Tennesse Red. The pairs of nucleolar chromosomes are heteromorphic to each other in the groundnut types studied.

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A CHROMOSOMAL CHIMERA IN *S. HALEPENSE* (LINN.)

By V. S. RAMAN and N. KRISHNASWAMY, Cytogenetics Laboratory, Agricultural College and Research Institute, Coimbatore

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(With plates II to IV)

In a programme of interspecific hybridisation of the grain and grass sorghums, a collection of forms of *Sorghum halepense* (Linn.), chiefly from Madras State has been maintained since 1947, at the Millets Breeding Station, Coimbatore. These have been transplanted several times without any changes being noticed. In January 1953, however, the collection from Humpi showed some branches with broader leaves which appeared to be vigorous. They were so markedly different as to draw particular attention and the clump was kept under observation. The tillers with the narrower leaves came to flower earlier and on examination were found to form 20 bivalents in the meiosis. The parental clump had been examined several times and had always given a tetraploid number of $2n=40$ chromosomes in the mitotic cells and 20 bivalents at meiosis. The tillers with broad leaves were somewhat late in flowering. These, on examining, were found to produce only ten bivalents, indicating a somatic number of $2n=20$. At this time, some naturally occurring diploid forms of *S. halepense* had been collected and planted nearby and a doubt arose if this diploid shoot could not be a stray seedling growing in the tetraploid clump. To verify this, the whole clump from Humpi was carefully lifted and the tillers with broad leaves were found to form part of the same clump as the tillers with the narrower leaves. This clearly proved that the plant in question was a diploid-tetraploid chimera.

The diploid portion was carefully cut out from the parental clump and transplanted apart. It soon recovered and produced a number of new shoots, of which again a good number bore narrow leaves and came to flower in 100 days (Plate II, figure 1 a and b). These were all found to be tetraploids with $2n=40$. The remaining tillers had broader leaves and came to flower in 250 days. The diploid and the tetraploid shoots were compared for differences in the phenotypical characters. The observations are summarised below.

Item	Parent clump	4n-tillers	2n-tillers
Pigmentation			
sheath, axil, } leaf-junction }	Reddish-purple	+	+
Leaf midrib	White	+	Slight, yellowish white
Stigma	Yellow	+	+

Item	Parent clump	4n-tillers	2n-tillers
Fresh anther	Yellow	+	+
Leaf :—(Plate III, figure 2) shape, colour.	Dark green, elliptic lanceolate	+	Yellowish green elliptic lanceolate
Length in cm.	84.95±0.15	90.7±0.07	91.0±0.07
Width in cm.	1.45±0.096	2.32±0.07	4.4±0.06
Node	Green, glabrous	+	+
Diameter of stem in cm.	0.8—1.0	0.8—1.2	1.4—1.8
Panicle (Plate III, figure 3)	Conical, very loose	+	+
Length of panicle	50 cm.	+	+
Sessile spikelets (Plate IV, figure 4)—length	0.5 cm.	+	+
width	0.2 cm.	+	+
Glume	Hairy, elliptic	+	+
Awn length	10—12 mm.	+	nil
Pollen fertility	94.8 per cent	+	+
Pollen size in μ	21.71±0.30	21.71±0.30	19.01±0.27
Plant height	300 cm.	+	+

It is seen from the above that there is no difference in the characters between the diploid and tetraploid shoots except in the leaf width, thicker stems and suppression of awns. The parent clump has so far not produced any broad leaved shoots again.

The flower buds from the diploid shoots were fixed in acetic alcohol (1 : 3) before separation of the clump. The mature sets of the diploid shoot were planted in sand and induced to root. The root tips were then fixed in Craf A and B solution. The counts in the somatic cells gave $2n=20$ chromosomes. This confirmed the observation in the acetocarmine smears. The complement was similar to that of the naturally occurring diploids. There was also not much difference evident between the karyotypes of the $2n$ and $4n$ -shoots. The meiosis was observed to be regular, forming ten bivalents with normal first and second divisions (Plate IV, figure 5 a, b and c). No case of any multivalent formation nor any disturbance in the meiosis was observed. Compared to the naturally occurring $2n$ -plants the meiosis varied in no details.

It becomes evident from these observations that a cell or cells in an axillary bud must have, owing to some cause, undergone somatic reduction giving rise to a tissue

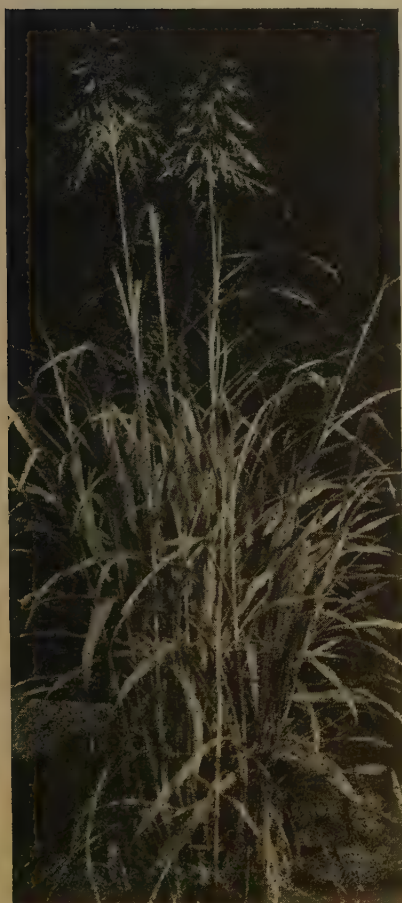


Fig. 1a. Chimera 2n-4n. whole plant.
4n-shoots in flower. 1/21 natural size



Fig. 1b. Transplanted chimerical portion; foreground
broad leaved 2n-shoot; back ground 4n-shoots with
nodal elongation 1/12 natural size



Fig. 2. Leaves
left-4n; right-2n.
1/7 natural size



Fig. 3. Panicles 1/5th natural size a-4n; b-2n

with reduced number of chromosomes. This reduction has been both quantitative and qualitative. *S. halepense* is supposed to be an allotetraploid (octoploid on the basic number five). Duara and Stebbins [1952] have concluded that it might be a segmental allopolyploid with the genomic make up $V_{12} V_{12} V_{13} V_{13}$. Based on the occurrence of natural diploids, Krishnaswamy and Raman [1953] have suggested it to be an autoallopolyploid. The genome is supposed to be made up of two sets of chromosomes each. Each one of the genomes is made up of one set of ten chromosomes closely homologous to that of the *Arundinaceae* group and the other of a wild plant also homologous but bringing with it the elliptic-lanceolate leaf shape and rhizomatous habit. Since, the diploid shoots in this chimera have remained essentially *halepense* in character and have not shown any exaggeration of characters of *Arundinaceae* group, it is to be concluded that the entire chromosome complement of the 4n-*halepense* has been reduced so that both complements are present in the diploid tillers. The only effects of diploidisation on the morphological characters are, (i) increase in the width of leaf and thickness of stem, (ii) lowering of the chlorophyll coloration of the leaf, (iii) suppression of awns and (iv) smaller pollen grains. These are characteristic of all the naturally occurring diploid forms collected from several different sources.

Somatic reduction has been recorded in a number of plants and also been artificially induced. Huskins [1948 and 1949] has reviewed the literature on this subject. Sodium ribose nucleate and sodium nucleate were found by him to induce somatic meiosis in root tips of *Allium cepa* and other plants. Such cells showing somatic meiosis were found by him also in onion bulbs which had become "flaccid after several months' storage". Huskins and Chouinard [1950] observed in *Rheo discolor* that "treatment with sodium nucleate increased the frequency of cells with somatic meiosis. Huskins and Chang [1950] found that cold treatment increased the proportion of cells of reductional type. The results have suggested to Huskins and his co-workers that "some type of repulsion between homologues or selective survival" was working in the induction of somatic meiosis. "Segregation may occur with or without chiasma formation. 'Distance conjugation' may occur in some plants." Patau [1950] also concluded that "the group separation is closely determined by what may be called, without physical implications, homologous repulsion". Hakansson [1950] noted in the root tips of species crosses chromosome elimination and variation as to the number between different roots and within the same root. Sampath [1950] has recorded cells with lower numbers of chromosomes in the root tip cells of a species cross in *Oryza*. Vaarma [1949] finds that the distribution of the chromosomes is completely random but that the chromosome groups thus formed are subject to a strong selection so that only suitable genome combinations give rise to viable super-reduced individuals. In the ovule mitoses he found the division of the tetraploid number into two equal similar groups as the most common. Brown [1947] is of the opinion that in cases where fertility is impaired by synaptic irregularities, meiosis acts as a regulating mechanism. Similarly, where sterility is due to chromosome unbalance or genic incompatibility, somatic eliminations might serve even in the absence of meiosis to establish a compatible chromosome

complement. Krug [1937] mentions spontaneous tetraploid shoots arising from an Octoploid in coffee. The branches differed in no way from normal *arabica* plants in fertility, flower and fruit characters. The seeds from these gave only *arabica* seedlings. He supposes quantitative as also qualitative equal reduction of chromosomes to have occurred. Watkins [1932] in analysing chromosomal chimeras has shown the probability that in cases where the haploid is less vigorous the diploid is more vigorous while where the haploid is vigorous the diploid becomes less vigorous.

These evidences show that there is every possibility in somatic meiosis for a perfect genomic separation to occur without in any way interfering with its fertility. In the chimera under consideration, the 2n-shoots are in no way less vigorous than the 4n. They have shown a tendency for a longer vegetative period similar to the naturally occurring 2n-forms. The suppression of awns on diploidisation is similar to the observations where, in induced autopolyploids awn is expressed in the tetraploids while it was absent in the diploids (*Oryza*, etc.). It can be expected that the progeny from the 2n-shoots will exhibit a high degree of homozygosity. A more detailed comparative study of these plants is being done and it is intended to employ the diploid obtained from the chimera in hybridisation studies also.

SUMMARY

The occurrence and characters of diploid shoots ($2n=20$) in a tetraploid clump ($2n=40$) of *Sorghum halepense* (Linn.) have been described. The origin is adduced to spontaneous somatic reduction of chromosomes in an axillary bud. The somatic meiosis has been both quantitatively and qualitatively equal. Some of the literature on somatic reduction of chromosomes has been reviewed.

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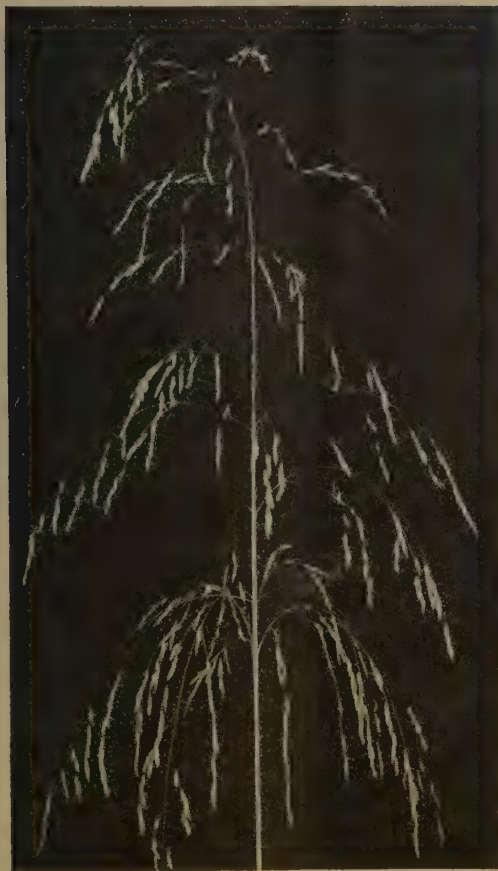


Fig. 4. Short branch $\times 2$. left-4n; right 2n

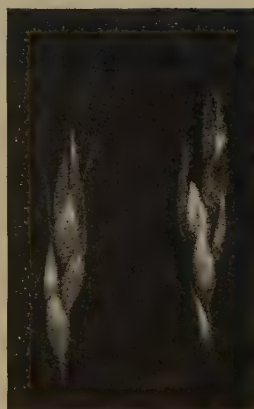


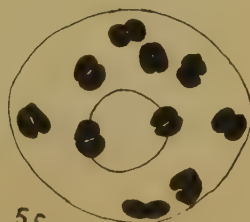
Fig. 5. $\times 3400a$ -somatic plate. 4n shoot; c-diakinesis is in 2n. flowers



5 a



5 b



5 c

INVESTIGATION ON THE PHYSIOLOGY OF GROWTH AND DEVELOPMENT OF *CROTALARIA JUNCEA* L. C. 12

By J. C. SEN GUPTA and SUMITRA TALUKDAR, Presidency College, Calcutta

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SINGH, Kapoor, and Choudhuri [1938] observed that *Crotalaria* can be induced to flower under 2 hours' light exposure. It flowered early when exposed to light periods which are short. From the point of view of fibre, those grown under 18 hours seemed to be the most suitable, as these plants were tall and unbranched. They came to flower about seven weeks later than controls.

The growth rate curves were characterised by a maximum at the approach of flowering and another at the seeding stage.

Singh and Choudhuri [1938] in another paper noted that *Crotalaria juncea* L. had a normal growth at 12 hours and that above this period the vegetative growth was much prolonged.

It has now been realised that, in order to introduce successfully improved method of cultivation with a view to obtaining the best growth and yield of crop plants, it is very useful to investigate the effects of environmental factors of nature and other factors on growth and development of the plants. In this connection, the effects of the varying environmental factors of nature are investigated by growing the plants in different season, and the most favourable time of sowing found out. Mention may be made here of the work of Ball and Holton on Egyptian cotton [1915], Gregory [1926] on the effect of climatic factors on the growth of barley, of Dastur and Singh [1944] and Dastur [1944] on cotton, Sen Gupta and Sen on jute [1944, 1948], and Sen Gupta and Mukherji on *mung* [1949].

The discovery by Lysenko in 1929, that presowing low or high temperature treatments can, to a great extent, influence the development of plants and induce early flowering, a phenomenon called vernalization, has been followed by several investigations and a very large number of crop plants have been experimented upon.

Since the discovery by Garner and Allard in 1918, that the daily period of light to which a plant is exposed may have a very great influence in the initiation of flowering (a phenomenon called photoperiodism), a very large number of investigations have been made on various crop plants on their photoperiodic effects. Sircar has recently reviewed the investigations on Vernalization and Photoperiodism in India [1948].

External factors, specially temperature, may have a remarkable influence on the photoperiodic effect, and the various investigations on different plants by different workers are recorded by Murneek [1948]; Sen and Pain [1948] have found that the photoperiodic effect is greatly influenced by season in *Sesamum*.

It has also been found with a variety of plants that a particular photoperiodic treatment conducive to flowering at the early stages of growth may be effective

in the induction of early flowering even under conditions of light period, which is not favourable for the induction of flowering, a phenomenon commonly known as photoperiodic induction, which seems to be true for both short and long day plants [Hamner, 1944].

This effect of pre-treatments by light periods is comparable to the effects of presowing temperature treatments, commonly known as vernalization, and recognition of this phenomenon has been of great value in detailed studies of photoperiodism in various plants and in an analysis of the nature of the photoperiodic reaction.

There is considerable evidence that the age of the plant determines, to a great extent, its sensitivity to the length of the day [Murneck, 1948 ; Hamner 1944].

From their observations of *Xanthium pennsylvanicum* plants grown in nutrient solutions of high or low NPK content, or with or without an external supply of N. Neidle [1939] and Naylor [1941] concluded that the amount of Nitrogen has no definite effect on photoperiodic response.

When the plants come to flower under a natural or artificial photoperiod, there takes place a change in the vegetative growth [Murneck, 1948]. It is sometimes seen that the stem elongation is retarded at the initiation of flower by suitable photoperiodic treatments, and this has been called photoperiodic inhibition [Murneck 1948]. Among apparently day natural plants, stem elongation has been found to be inhibited by long day in cucumber [Danielson, 1944] and by short day in some tobaccos [Dennison, 1945].

Synthetic hormones have been applied to seeds in numerous investigations with a view to increase the percentage and rate of germination, accelerate growth rate and advance the date of maturity, increase the yield of crops and to counteract deleterious effects of fungicides. Although more negative results have been obtained than positive, Amlong and Naundorf [1939] reported significant success with Alfalfa, and sugar beet by treatments with potassium salts of indole acetic acid, indole butyric acid, and Naphthalene acetic acid.

The present investigation was undertaken to understand the nature of response in the (i) growth and development of the plants to the variations of climatic factors of nature with reference to the different sowing times, (ii) to presowing temperature treatments with or without the addition of the hormone indole acetic acid, (iii) to different photoperiodic treatments.

The photoperiodic behaviour was investigated in the following details :

- (a) The effect of continuous photoperiodic treatments till flowering ;
- (b) The relative influence of other environmental factors, chiefly temperature of the different seasons ;
- (c) The effect of light period treatments for different durations at the early stage to study induction effects ;
- (d) The effect of the age of the plant on the photoperiodic response ;
- (e) The influence of manuring the plants with ammonium sulphate and acid potassium phosphate on the photoperiodic response, and also to find out
- (f) the influence of different photoperiodic treatments on the vegetative growth in relation to the onset of the reproductive phase.

EXPERIMENTAL PROCEDURE

Pure strain seeds of *Crotalaria juncea* Linn. C.12 obtained through the courtesy of State Agricultural Department, West Bengal, have been used in this investigation.

Seeds selected to be of uniform size as far as possible were sown in earthenware pots of 13 in. diameter at the top and 13 in. height with a capacity of 18 litres, in well manured garden soil at the experimental garden of the Presidency College, Calcutta. The seeds were sown in a ring equidistant from the rim and the centre and as the seedlings grew, only four plants more or less equidistant from each other were kept. There were three pots, i.e. 12 plants per treatment. In the case of presowing temperature treatments, sowing was done only in plots.

The nature of treatments and the readings taken are given below :

(i) *Time of sowing experiments*

Seeds were sown once every month on the 21st, beginning from 21-4-51, and there were twelve sowings. Seeds were sown in earthenware pots of 13 in. diameter at the top and 13 in. height, in the manner mentioned before. Four plants more or less at the same distance were kept per pot, and there were three pots per sowing, i.e. 12 plants per treatment.

As the plants grew, fortnightly readings on height and number of internodes were continued till there was no further appreciable growth, and the plants showed signs of withering and the leaves were discoloured, most of them being shed. But the height and number of internodes, as recorded in the fortnightly reading just after flowering, are included in a separate table, together with the flowering time.

(ii) *Temperature treatments*

Prior to subjecting the seeds to temperature treatment, the time required for soaking in water which would induce just sprouting of the seeds was determined by experiments and found to be two hours. The seeds were then soaked for the requisite period and were placed in an electrically operated constant temperature bath kept at 30—35°C for the high temperature treatments, and in a refrigerator at a temperature of 2—4°C for the low temperature treatments. The seeds were placed over a moist filter paper in a petridish and water was added to the filter paper from time to time to keep the seeds in a moist condition, in the high temperature bath. In the high temperature treatment, the sprouting seeds of Sunn-hemp were very susceptible to fungal attack, and it was not possible to treat them for more than seven days.

A beaker with KOH solution was kept in the chamber to absorb the CO₂ of the respiration of seeds. The low temperature treatment was for periods of 26, 14 and 7 days. Some of the seeds at the low temperature treatment were continuously treated in petridishes with a dilute solution of Indole acetic acid at a concentration of 1 ppm for a period of 30 days. Other seeds receiving cold temperature treatment without hormone were kept in separate funnels lined with moist filter paper and glass wool, on different conical flasks containing dilute KOH solution to absorb the CO₂ of respiration.

Seeds were sown on the 21st July to investigate further the effects of Indole acetic acid and cold temperature and for 30 days the treatment continued ; a warm temperature treatment of seven days was also followed up. The seeds of all these treatments were sown in plots of 6 ft. \times 3 ft. separate plots being used for the sowing of seeds of different treatments. The seeds were sown in rows 6 in. apart, so that there were six rows in each plot. Twelve plants were selected by the random method for each treatment from the plot.

In every case, in addition to the flowering and the fruiting time, the following readings were taken at the flowering stage, to ascertain the amount of vegetative growth at this stage :

- (1) Total height of the main stem in c.m.
- (2) The number of internodes.
- (3) The circumference of the stem at the base at flowering.

These readings were taken separately for each plant and expressed as mean.

(iii) *Photoperiodic treatments*

The initial procedure was the same for all the five photoperiodic experiments. Sowing was done in pots of uniform size and 12 plants in three pots were taken for every treatment.

(a) Continuous photoperiodic treatment : of 8 hours, 10 hours, 12 hours, 14 hours and 16 hours, were given until flowering. For light periods shorter than the natural day length, the pots were removed to a ventilated dark chamber at appropriate times and allowed to remain there until dusk. The extra light was given from 100 C.P. electric bulb at a distance of 1 metre. For this experiment, seeds were sown on the 21st May, 1951.

(b) Photoperiodism at different seasons : to investigate the influence of the environmental factors of the different seasons, seeds were sown on 21-5-51, 21-8-51, 21-11-51 and 21-2-52 and the plants subjected to photoperiodic treatments of 10, 12 and 14 hours till the initiation of flowering.

(c) Photoperiodic induction : for the study of the induction effect, seeds were sown on 21-8-51 and the plants treated with daily light periods of 10, 12 and 14 hours, for 14 days in one set and for 28 days in another and subsequently kept in the open, exposed to natural day lengths.

(d) Photoperiodism at different ages : for a study of the effect of the age of the plant at which the photoperiodic treatment is applied, seeds were sown on 21-5-51 and allowed to grow under natural conditions and treatment with daily light periods of 10, 12 and 14 hours commenced at the age of 30 days in one set and 45 days in the second. Treatments were continued till the initiation of flowering.

(e) Photoperiodism and fertilizers : to investigate how far addition of inorganic manures influences the photoperiodic response, the plants were manured with ammonium sulphate in one set and acid potassium phosphate in another. Each pot received 10 grams of the manure, 5 grams being added when the plants were 10 days' old and the second 5 grams when they were 20 days' old. Treatments at 10, 12 and 14 hours commenced when the plants were 30 days' old in each case.

A control set, exposed to the natural conditions of environment, was kept.

TABLE I
Effect of time of sowing

Date of sowing	Height in cm.	S. E.	Group No.	Per day increase in height	No. of internodes	S. E.	Group No.	Average length of each internode	Circumference at the base of the stem at flowering	S. E.	Group No.	Flower time in days	Fruiting time in days
21-4-51	307.5	8.23	I	1.86	189.0	1.23	I	1.62	5.8	0.063	I	156	163
21-5-51	296.1	6.51	II	1.80	157.0	2.31	II	1.88	5.9	0.854	I	112	120
21-6-51	204.7	5.91	III	1.59	122.4	0.752	III	1.67	5.1	0.234	I	72	80
21-7-51	183.1	2.95	IV	1.56	99.9	3.46	IV	1.58	2.2	0.089	II	74	83
21-8-51	101.5	5.35	VI	1.00	77.4	0.852	V	1.31	1.4	0.123	II	30	39
21-9-51	74.4	6.21	VII	1.03	60.0	1.59	VI	1.24	1.0	0.356	III	23	31
21-10-51	65.1	2.95	VIII	1.86	58.2	0.831	VI	1.11	0.93	0.035	III	20	30
21-11-51	69.1	1.86	VII & VIII	2.03	35.0	0.459	VIII	1.97	1.1	0.121	II	25	33
21-12-51	50.0	2.86	IX	1.42	34.0	0.392	VIII	1.47	1.2	0.113	II	25	33
21-1-52	50.8	1.25	IX	0.86	40.3	0.112	VII	1.24	0.92	0.086	III	23	31
21-2-52	46.3	3.29	IX	1.10	44.6	0.865	VII	1.04	1.2	0.542	II	20	30
21-3-52	117.4	5.23	V	0.99	80.0	0.531	V	1.46	1.7	0.071	II	118	Not noted

Statistical difference in means at 5 per cent level of significance is indicated by different Group Numbers, while means having the same Group Number are not significantly different from one another. Border line cases are indicated by more than one Group Number. This convention has been followed in all the Tables.

The following readings were taken at the flowering stage to know about how far the vegetative growth is influenced by the photoperiodic treatments, in addition to the flowering and fruiting times :

- (1) Total height of the main stem in cm.
- (2) Number of internodes.
- (3) Circumference of the stem at the base, at flowering.

As usual, these readings were taken separately for each plant and expressed as mean.

TABLE II(a)

Effect of presowing high and low temperature treatments on the flowering time and height at flowering

Sown on 21-5-51

Treatment	Flowering time in days	S. E.	Group No.	Mean height at flowering in cm.	S. E.	Group No.	Fruiting time in days
30-35°C for 7 days	112.0	1.01	I	254.5	4.58	I	127
2-4°C for 26 days	114.2	1.89	I	210.1	5.76	II	131
2-4°C for 14 days	112.2	1.39	I	210.7	4.31	II	128
2-4°C for 7 days	111.0	1.05	I	200.2	2.39	III	8
Control	109.1	1.32	II	207.7	3.96	II	125

TABLE II(b)

Effect of presowing temperature treatment with and without the addition of 1 ppm. indole acetic acid on the flowering time and height at flowering

Sown on 21-7-51

Treatment	Flowering time in days	S.E.	Group No.	Mean height in cm. at flowering	S. E.	Group No.	Fruiting time in days
2-4°C for 30 days	80.0	0.018	I	118.3	5.75	II	89
2-4°C 1AA for 30 days	82.5	1.21	I	92.0	3.25	III	90
30-35°C for 7 days	81.0	0.057	I	75.2	2.95	IV	91
Control	74.0	0.091	II	125.0	5.72	I	84

TABLE III (a)

Effect of different photoperiods on the flowering time and height at flowering at the normal growing season

Sown on 21-5-51

Treatments	Flowering time in days	S. E.	Group No.	Mean height in cm. at flowering	S. E.	Group No.
8 hours	73.3	0.023	III	52.3	0.858	IV
10 hours	37.4	0.982	IV	39.0	3.25	V
12 hours	33.4	0.085	IV	35.3	1.23	V
14 hours	131.0	0.098	I	226.5	3.17	I
16 hours	No flowering			153.7*	3.58	III
Control	109.0	2.91	II	215.7	4.23	II

*Height recorded on the 206th day

TABLE III (b)

Effect of climatic factors of different seasons on photoperiodic behaviour

CONTROL							
Sowing date	Mean flowering time in days	S. E.	Group No.	Mean height in cm.	S. E.	Group No.	Mean fruiting time in days
21-5-51	109	2.91	I	215.7	4.23	I	125
21-8-51	30.4	0.054	II	70.1	2.55	II	37
21-11-51	25.4	0.098	III	53.7	1.35	III	29
21-2-52	20.0	0.023	IV	19.7	0.988	IV	28

10 HOURS

21-5-51	37.4	0.982	II	39.0	8.25	III	43
21-8-51	46.4	1.01	I	91.9	2.15	I	50
21-11-51	29.2	0.056	III	69.7	1.25	II	39
21-2-52	29.0	0.085	III	20.6	1.35	IV	38

TABLE III (b)—*contd.*

Effect of climatic factors of different seasons on photoperiodic behaviour

12 HOURS

Sowing date	Mean flowering time in days	S. E.	Group No.	Mean height in cm.	S. E.	Group No.	Mean fruiting time in days
21-5-51	33.4	0.085	I	35.3	1.23	III	45
21-8-51	33.6	1.56	I	93.0	2.76	I	56
21-11-51	30.6	0.082	II	70.5	1.12	II	39
21-2-52	29.8	0.055	II	80.6	1.76	IV	36

14 HOURS

21-5-51	131.0	0.098	I	226.5	3.17	I	143
21-8-51	76.0	1.37	II	144.9	4.15	II	85
21-11-51	73.0	2.01	II	106.7	4.59	III	80
21-2-52	Died.						

TABLE III (c)

Effect of photoperiodic induction for 14 and 28 days

Sown on 21-8-51

Light period	Duration	Mean flowering time in days	S. E.	Group No.	Height in cm. at flowering	S. E.	Group No.
10 hours	Continuous	46.4	1.01	III	91.9	2.15	III
	For 14 days	23.0	0.542	VI	68.0	1.34	IV
	For 28 days	30.8	0.025	VI	71.2	1.87	IV
12 hours	Continuous	33.6	1.56	V and VI	93.0	2.76	III
	For 14 days	35.0	0.042	V	68.5	1.59	IV
	For 28 days	41.7	0.051	IV	54.6	1.89	V
14 hours	Continuous	76.0	1.37	I	144.9	4.15	I
	For 14 days	45.0	0.098	III	71.3	2.01	IV
	For 28 days	49.5	0.852	II	97.6	3.21	II
Control	..	80.4	0.054	VI	70.1	2.55	IV

TABLE III (d)

Effect of application of ammonium sulphate and potassium hydrogen phosphate on the flowering time and height at flowering

Sown on 21-5-51

Light period	Treatment at the age of	Manure applied	Mean flowering time in days	S. E.	Group No.	Height in cm. at flowering	S. E.	Group No.
10 hours	Continuous	..	37.4	0.082	IV	39.0	3.25	IV
	30 days	Ammonium sulphate	47.0	1.57	III	103.3	4.03	II
	30 days	Potassium phosphate	47.5	0.876	III	99.7	4.53	II
	30 days	None	47.0	1.22	III	93.0	3.79	II
12 hours	Continuous	..	33.4	0.085	IV	35.3	1.23	IV
	30 days	Ammonium sulphate	46.2	0.083	III	93.4	2.59	III
	30 days	Potassium phosphate	52.6	1.97	II	104.4	3.01	II
	30 days	None	44.2	0.001	III	100.3	3.98	II
14 hours	Continuous	..	181.4	0.098	I	226.5	3.17	I
	30 days	Ammonium sulphate	131.0	2.05	I	223.1	3.53	I
	30 days	Potassium phosphate	131.0	0.001	I	231.0	3.25	I
	30 days	None	133.0	0.020	I	230.5	2.98	I

TABLE III (e)

Effect of age on the photoperiodic behaviour

Sown on 21-5-51

Light period	Treatment at the age of	Mean flowering time in days	S. E.	Group No.	Height in cm. at flowering	S. E.	Group No.
10 hours	Continuous	37.4	0.082	VI	39.0	3.25	VII
	30 days	47.0	1.22	V	93.0	3.79	VI
	45 days	61.1	1.23	IV	143.9	1.99	V
12 hours	Continuous	33.4	0.085	VI	35.3	1.23	VII
	30 days	44.2	0.001	V	100.3	3.98	VI
	45 days	77.1	0.875	III	161.6	1.36	IV
14 hours	Continuous	131.0	0.098	I	226.3	3.17	II
	30 days	133.0	0.020	I	230.3	2.98	II
	45 days	132.0	0.037	I	244.2	4.56	I
Control	(Normal)	109.0	2.91	II	215.7	4.23	III

METEOROLOGICAL DATA

The maximum, minimum and mean temperature, the rainfall, relative humidity and total hours of bright sunshine in fortnightly means, and the daily light period on the dates mentioned are given in Table IV.

TABLE IV

Meteorological data

Date	Total hours of bright sunshine	Max. Temp. (°F)	Min. Temp. (°F)	Mean Temp. (°F)	Rainfall (inches)	Relative Humidity	Daily light period
21-4-51	10-3	95	76	87-5	0-00	76	12-48
5-5-51	10-02	98-16	86-04	92-1	0-126	60-482	13-2
19-5-51	10-4	101-60	97-12	99-36	0-00143	64-616	13-17
2-6-51	10-08	96-06	88-26	92-61	0-086	59-907	13-26
16-6-51	4-6	92-02	86-08	89-02	0-325	74-79	13-30
30-6-51	4-18	96-16	86-19	91-17	0-354	75-637	13-30
14-7-51	3-54	89-32	83-49	86-40	0-32	88-776	13-25
28-7-51	6-14	87-24	88-5	86-87	0-32	79-84	13-14
11-8-51	6-06	90-68	85-44	88-06	0-24	83-177	13-0
25-8-51	4-25	96-17	85-31	90-74	0-43	82-02	12-46
8-9-51	4-9	89-17	84-8	86-98	0-32	89-047	12-26
22-9-51	5-91	92-44	85-23	88-83	0-017	72-11	12-8
6-10-51	7-04	90-02	78-97	84-49	0-339	68-377	11-51
20-10-51	3-17	91-442	79-36	85-40	0-1269	75-11	11-34
3-11-51	7-98	88-58	75-54	82-06	0-0434	64-29	11-17
17-11-51	7-74	86-86	67-83	77-34	0-000	66-68	11-2
1-12-51	7-83	80-92	61-23	71-07	0-14	56-44	10-51
15-12-51	8-54	82-05	62-74	72-39	0-000	71-11	10-46
29-12-51	8-75	79-15	55-75	67-45	0-000	59-76	10-46
12-1-52	5-91	81-23	56-25	68-76	0-00	57-42	10-51
26-1-52	8-64	81-29	56-3	68-79	0-00	53-24	11-3
9-2-52	7-76	84-67	61-66	73-16	0-00	38-6	11-17
23-2-52	8-22	92-52	65-2	78-86	0-0023	35-618	11-34
8-3-52	5-7	88-75	67-93	78-34	0-005	58-11	11-50
22-3-52	6-56	91-22	65-51	78-36	0-081	53-52	12-3
5-4-52	8-18	92-34	77-09	84-71	0-021	64-47	12-26
19-4-52	9-64	94-68	74-92	84-80	0-09	46-86	12-45
3-5-52	9-8	92-32	81-57	86-94	0-25	59-82	13-0

DISCUSSION

Growth and development of Sunn hemp C.12 are greatly influenced by the sowing time. From the sowing of 21st April, the height and number of internodes gradually decrease with lateness of sowing till the sowing of March where an increase in height from the previous sowing of February is observed, and the height is intermediate between the sowings of July and August. The average increase in height is seen to be highest in the November sowing, but here the vegetative period is short. Between the four sowings of April, May, June and July, which show the greater heights than the rest, the average per day increase in height is not widely different from one another, and the circumference at the base of the stem of the first three sowings are also not significantly different from one another. It should be noted that the plants, sown during the months from April to September, all flower between the first September and the fourteenth October, i.e. for a difference in sowing time of 150 days, the flowering time falls within a period of 44 days, which indicate that the environmental factors of the month of September upto the middle of October are favourable for the onset of the reproductive phase. During this period the maximum temperature varies between 83°F and 92°F and the minimum temperature between 75°F and 80°F and the daily light period between 11 hours 42 minutes and 12 hours 36 minutes. The sowings of October to February have very short vegetative periods and poor vegetative growth. The sowings of August and September also have short vegetative periods, but the vegetative growth is slightly better in these sowings, especially in the sowing of August.

The plants sown in April and May are exposed to comparatively high temperature (maximum between 107.1°F and 93°F and minimum between 84.6°F and 71.3°F) and long daily hours of bright sunshine and are exposed to high temperature and rainy season of the monsoon after they are one and two months old. The plants sown in June and July are also exposed to high temperature, but they are exposed to the rainy season and comparatively lower daily hours of bright sunshine from the very beginning. The plants sown in August and September grow under similar conditions, but at slightly lower temperature. The plants sown in October to February grow under much lower temperature conditions and have very poor growth. In general, the vegetative periods of the plants sown in August to February are very low. The sowings in March, however, again grow under longer hours of bright sunshine and higher temperature and have much longer vegetative growth than the sowings of previous months.

Best growth is thus found in plants sown in April and May, which grow under higher temperature and longer daily hours of bright sunshine, followed by the rainy season, which is very favourable for the vegetative growth of this plant.

It may be concluded that the sowing time most favourable for vegetative growth is between the months of April and May under the environmental conditions prevailing here.

The presowing temperature treatments (Table IIa) produce significant lateness of flowering from the control and this plant is thus similar in behaviour to *H. Sabdariffa* L.N.P.5 and several other plants found by others, like Ivanov on Flax [1930],

Sircar [1945] and Kar and Adhikary [1945] on rice and Kar [1940, 42-43], Pal and Murty [1941] on wheat. There is no significant difference between the different low and the only high temperature treatments. The growth in height is, however, greatest in 30-35°C treatment for seven days and lowest in 2-4°C treatment for seven days and there is no significant difference between the rest.

In the experiment with the addition of 1 ppm. indole acetic acid (Table IIb), there is no difference in flowering time between the low temperature treatment with and without indole acetic acid and high temperature treatment for seven days. The height is, however, greatest in the control and lowest in seven days' high temperature treatment, and between the two low temperature treatments at 30 days, the treatment, without the addition of indole acetic acid, has significantly greater growth than the one with the addition of the hormone. It should, however, be noted that both the flowering time and height are greater in all the treatments of the sowing on 21st May than in the sowing of 21st July.

It has been found by the experiment on the photoperiodic effect that the flowering is considerably enhanced by the 10 hours and 12 hours treatments and is also much earlier in 8 hours treatment than the control and the flowering is considerably delayed in the 14 hours treatment and there is no flowering in 16 hours treatment. The plant is thus clearly a short day plant and there is an earliness of 72 days in the 10 hours and of 76 days in 12 hours from the control, the differences between 10 hours and 12 hours being statistically insignificant. The heights at flowering follow the same sequence, and the vegetative growth is considerably inhibited in the 12 hours, 10 hours and 8 hours treatment and accelerated in the long day treatment of 14 hours. The critical light period seems to lie between 10 hours and 12 hours which is in conformity with the finding of Singh and Choudhri [1938]. Plants induced to flower under the light periods of 12 and 10 hours have a marked development of branches from below the apex at the flowering stage. This is less marked in the controls and the 14 and 16 hours groups. The development of branches at the flowering stage in this plant reminds one of that of jute as recorded by Sen Gupta and Sen [1944], and of *Crotalaria* as noted by Singh and Choudhuri [1938].

The photoperiodic effect (Table IIIb) appears to be influenced by the climatic factors of the different seasons. In the control and 14 hours treatments the flowering time and height at flowering decrease with lateness of sowing, commencing on the sowing of May, although there is no significant differences between the heights at 14 hours treatments of the May and August sowing. The photoperiodic effect in 10 hours and 12 hours treatments follows a different course. The flowering time is longer in the sowing in August than the same in May in 10 hours treatment, and in 12 hours treatment there is no significant difference between the two sowings. Between the sowings of November and February, the difference in the flowering time is not significant, both in 10 hours and 12 hours treatments. What is very remarkable is the fact that in the three off-season sowings of August, November and February, the plants in the normal control set flower earlier than in the 10 hours and 12 hours treatment; apparently in these seasons the plant does not behave as a short day plant. The influence of the variation of the climatic conditions is

thus very great. Temperature is known to influence the photoperiodic behaviour greatly. Each species seems to have its own temperature range for optional flowering response, and the temperature requirement during the light period may be quite different from requirements during the dark period (Leopold 1951). Some plants, with a photoperiodic requirement for flowering, can be modified in that requirement or, in some cases, the photoperiods can be entirely substituted for, by a temperature change. (Lang and Melchers, 1943 ; Murneek, 1940.) In this plant the vegetative growth becomes very poor in the sowings of August, November, and February, which is due evidently to the comparatively lower temperature more than any other factor. It is thus clear that Sunn hemp C.12 requires the temperature ranges to which the sowing of May is exposed to maintain its short day character, and response to photoperiods.

The plants for the experiment on photoperiodic induction (Table IIIc) were sown in August and, as mentioned before, in all the continuous treatments the flowering was delayed from the control. In 10 hours treatment, there was a clear induction effect for both 14 days and 28 days treatments in which the flowering took place earlier and growth in height was inhibited. It appears, under the environmental conditions of the experiment, that the inner conditions for the response to the photoperiodic stimulus are more clearly developed in shorter periods of 14 days and 28 days than when the plants are exposed to continuous treatment.

Addition of Ammonium sulphate and Potassium phosphate (Table IIIId) has no influence on the photoperiodic effect. It may be mentioned that Sen Gupta and Sen [1947] found no effect of Ammonium sulphate on the flowering in jute (*C. olitorius* L.) and this supports the conclusions of Neidle [1939] and Naylor [1941]. The vegetative growth is, however, promoted by the addition of the manures.

The age of the plant at which treatment is applied (Table IIIe) is known to influence the photoperiodic response. In Sunn hemp C.12, it is found that in all cases the flowering takes place earlier after the commencement of treatment when it is applied at 30 days and 45 days than in the continuous treatment. When the plants have grown under natural condition for 30 or 45 days, on change over to favourable photo inductive cycles, they develop the internal conditions for photoperiodic response, whether by way of production or conversion into or transport of flower producing substances, or by other unknown means, more quickly.

SUMMARY

1. *Crotalaria juncea* L. (Sunn hemp C.12) was grown in pots at intervals of one month for 12 months, commencing from the 21st April in 1951-52, there being thus 12 sowings.

2. The vegetative growth and development were greatly influenced by the time of sowing.

3. For widely different sowing times between April and September (150 days), the flowering took place within the month of September upto the middle of October

(44 days); the vegetative period thus became progressively shorter with the lateness of sowing. The significance of the photoperiodic effect is discussed in this connection.

4. The growth in height, number of internodes and the circumference at the base of the stem became progressively shorter with the lateness of sowing, the sowings of October to February showing very poor growth, which is ascribed to the comparatively lower temperature prevailing during these months.

5. The probable effects of the different environmental factors on the growth and development of the species have been discussed.

6. A favourable time for sowing Sunn hemp C.12 under the environmental factors prevailing here seems to be between the months of April and May.

7. In another set of experiments, seeds were vernalized at low and high temperature for different periods and, in one experiment, 1 ppm. indole acetic acid was applied to the low temperature treatment.

8. In every case, the presowing temperature treatments produced distinct lateness in flowering from control, but there was no significant difference between the treatments themselves.

9. The addition of the hormone indole acetic acid was without effect on the flowering response.

10. In a third series of experiments, the photoperiodic behaviour was investigated by:

- (a) 8 hours, 12 hours, 14 hours and 16 hours treatments at the normal growing season ;
- (b) 10 hours, 12 hours, and 14 hours treatments at four growing seasons at intervals of 3 months for the effect of climatic factors of the different season on the photoperiodic behaviour ;
- (c) 10 hours, 12 hours and 14 hours treatment for 14 and 28 days at the early stages for the induction effect ;
- (d) treatments at 10 hours, 12 hours and 14 hours at the age of 30 days after manuring the plants with Ammonium sulphate and Potassium hydrogen phosphate for the effect of the manures ; and
- (e) 10 hours, 12 hours and 14 hours treatments at the ages of 30 and 45 days for the effect of age.

11. The plant shows distinct photoperiodic response :

- (a) Sunn hemp C.12 has been found to be a short day plant ;
- (b) the climatic factors of the different seasons influence the photoperiodic behaviour, and the effect is very remarkable in the 10 hours and 12 hours treatments ; the probable effect of temperature is discussed in this connection ;
- (c) there was a clear induction effect ;

- (d) addition of Ammonium sulphate and Potassium hydrogen phosphate has no effect on the photoperiodic response;
- (e) the plants respond earlier after the commencement of treatment when they are 30 and 45 days old.

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STUDIES ON INDIAN CLOVER (*SENJI*) IN THE PUNJAB

By H. C. MALIK, Economic Botanist (Fodder), Punjab, Sirsa,
District Hissar

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INDIAN clover or *senji* (*Melilotus parviflora*) is one of the outstanding legumes grown exclusively for forage during winter season in the Punjab. In fact, prior to the introduction of berseem, it was the chief legume which occupied a very important position as a catch crop for fodder [F. B., 1950]. It is also highly soil restorative and has no equal as such [Kartar Singh, 1947]. With the increasing popularity of berseem, however, *senji* has been replaced by the former to a considerable extent. As a matter of fact *senji* is now primarily grown under conditions where either it is difficult to raise a berseem crop because of the limited supply of irrigation water, or where it is desired to fit in the subsequent crop in the rotation, as for example sugarcane and cotton. Even then it occupies a considerable acreage, especially in the well irrigated areas of Jullundur, Hoshiarpur, Ludhiana and Ambala districts. According to the census of fodder crop taken in the Punjab in 1937-38, *senji* occupied an acreage of three lakh in the districts now in the Punjab (India); this constituted about 20 per cent of the total area under fodder crops in this State. It is, however, on the decrease as will be seen from the figures of acreage for the quinquennium ending 1948-49.

	Acres
1944-45	1,33,971
1945-46	1,47,329
1946-47	1,44,719
1947-48	1,18,336
1948-49	79,881

Not only, Indian clover (*senji*) is a highly sustaining forage having a fairly narrow nutritive ratio of 4.1, as compared to 3.6 of berseem and 2.6 of lucerne [Sen, 1946], but also its easy cultivation and suitability to a wide range of soil, climatic conditions and rotations, are responsible for making it a popular forage with the farmer. Some of the common rotations reported by Kartar Singh [1947] in which *senji* is grown are given below:

1. Wheat-maize-*senji*-cotton
2. Maize-*senji*-cotton
3. Wheat-maize-*senji*-sugarcane
4. Wheat-cotton-*senji*

In view of its importance in the farm economy, work on its improvement with a view to evolving varieties that would give increased yield under the Punjab conditions, has been under way at the Fodder Research Station, Sirsa, and as a result a variety named *senji* No. 1 was evolved and approved for distribution in this State by the Department of Agriculture, Punjab.

REVIEW OF LITERATURE

Of the several kinds known, only yellow blossomed (*Melilotus parviflora*) is commonly grown in this State. The white flowered (*Melilotus elba*) a late maturing variety is found growing wild or in slight admixture with the former. Definite attempts at its improvement are of recent origin, and high yielding forage varieties have been developed on the basis of growth and maturity.

Garver, *et al.*, [1943] described 15 varieties of sweet clover as grown in Nebraska on the basis of the diversity in growth, maturity, coarseness of stem and seedling vigour. According to the same author definite breeding work has been under way in Canada under the direction of L. E. Kirk, with a view to evolving winter hardy, fine stemmed, leafy, disease resistant and more palatable sorts by isolation of desirable forms through selection within inbred lines.

Sweet clover breeding has also been carried out in Russia at the Black Soil Regional Plant Breeding Centre at the Maikop and Detsko-Seto stations in the North Caucasus and at Sorator by growing commercial varieties and native wild strains.

Many thousands of plants from different sources were grown and studied at the Kaiser Wilhelum Institute in Germany from which 51 individuals were selected.

Less attention was paid to the selection of varieties from yellow sweet clover than from white, because the former was found to be less productive and much earlier than those of white sweet clover. Even varieties of sweet clover developed in the U. S. A., as for example, Albert, Madison country and St. Louis Valley were much like the commercial yellow.

MATERIAL AND METHODS

Improvement of Indian clover has been principally concerned with the study of the behaviour and growth of a very large number of samples collected from different areas of the State as well as with the number of introductions from abroad. The latter did not succeed well and observation and experimentation with the local material were resorted to.

Study of more than 500 single plants was made from among the samples with regard to their flowering, leaf, size and period taken to become ready for harvest as forage. On the basis of these characters a number of selections were made and compared for their forage value, firstly in the progeny row tests and later on in regular experiments according to the randomized system of field trials for 12 years from 1935-36 to 1946-47 at the Fodder Research Station, Sirsa, as well as at the Agricultural Experimental Stations in the State. The continuity of trials at the Fodder Research Station, Sirsa, and at the various Departmental Agricultural Stations could not be maintained because of the administrative changes at the places concerned during that period. Later on *senji* No. 1 which appeared superior in forage value to all other selections was tested at the *zamindara* lands also.

Senji No. 1 is a tall growing, yellow flowered variety which on an average takes 9-10 days more to flower than local. It has broad leaves and gives out a large number of branches resulting in the plant having a bushy appearance.

RESULTS AND DISCUSSION

The yield of forage obtained from the selections as compared to that of the local, and their critical differences are given in Table I.

TABLE I

The forage yield of selections of Indian clover (senji) at the Fodder Research Station, Sirsa

Year	Yield per acre in maunds		Difference from local	C. D. per acre in maunds at		Significance
	No. 1	Local		0-05	0-01	
1935-36	202.4	145.2	56.2	39.7	72.9	Significant
1936-37	286.9	229.8	57.1	16.0	23.8	
1939-40	484.0	407.0	77.0	45.0	68.2	
1940-41	309.0	289.5	19.5	10.4	16.4	
1941-42	358.0	301.0	57.0	33.2	47.2	

Table I shows the yields of green forage of *Senji* No. 1 and local at the Fodder Research Station, Sirsa, for five years. Though forage yields showed a good deal of variation from one year to the other in the five tests, on an average an out-turn of 328 maunds was obtained from the new variety compared to 247.7 maunds per acre from local, showing a difference of 53.3 maunds per acre or 19.3 per cent higher than the control.

The tests were not only conducted at the Fodder Research Station but also at the Departmental Agricultural Stations representing different soil and climatic conditions of the State. The results of these tests are given in Table II.

TABLE II

The forage yield of senji at different Agricultural Experimental Stations

Station	Year	Yield per acre in maunds		Difference from local	C. D. per acre in maunds		Significance
		No. 1	Local		0-05	0-01	
Gurdaspur	1940-41	210.0	196.0	14.0	8.4	12.5	Significant
	1941-42	226.0	211.0	15.0	Not significant
	1945-46	269.5	258.7	10.8	8.1	11.2	Significant
	1946-47	312.0	303.1	8.9	17.3	25.6	Not significant
Average		254.3	242.2	12.1			

TABLE II—(contd.)

The forage yield of senji at different Agricultural Experiment Stations

Station	Year	Yield per acre in maunds		Difference from local	C ₆ D. per acre in maunds		Significance
		No. 1	Local		0.05	0.01	
Jullundur	1940-41	355.5	341.2	14.3	Not significant
	1941-42	488.7	456.2	32.5	19.7	27.3	Significant
	1945-46	341.1	329.9	11.2	Not significant
	1946-47	403.5	375.0	28.5	Not significant
Average		402.2	380.5	21.7			
Hansi	1941-42	125.0	121.0	4.0	Not significant
	1945-46	343.4	355.2	-11.8	Not significant
	1946-47	308.8	301.9	6.9	16.9	24.2	Not significant
Average		259.0	259.3	-0.3			
Lyallpur	1941-42	348.7	325.6	23.1	16.7	26.2	Significant
	1945-46	216.7	211.6	5.1	Not significant
	1946-47	359.9	332.4	27.5	19.3	27.2	Significant
Average		308.2	289.8	18.6			
Montgomery	1940-41	255.0	187.0	68.0	26.2	41.1	Significant
	1941-42	361.2	328.7	32.5	14.8	23.2	Significant
	1945-46	216.7	211.6	5.1	Not significant
Average		356.6	294.1	62.5			
Multan	1940-41	180.0	164.0	16.0	Not significant
	1946-47	295.2	229.0	66.2	14.1	22.2	Significant
Average		237.6	196.5	41.1			

The results of trials carried out at the departmental farms revealed the superiority of No. 1 over the control. Out of the 19 tests in favour of No. 1, yields were significantly higher in nine cases, and higher but not significant in ten. Taking results of the various tests at the different departmental farms separately, it was evident that No. 1 was superior to local at Gurdaspur, Jullundur, Lyallpur, Montgomery and Multan giving on an average 254, 402, 308, 356 and 237 maunds compared

to 242, 380, 289, 294 and 196 maunds per acre from local respectively. Yields of No. 1 and local were almost equal, i.e. 259 maunds per acre at the Hansi Agricultural Station.

Besides experiments at the Fodder Research Station, Sirsa and the departmental farm, No. 1 was compared with local at the farmers lands in the State. The yields of forage obtained at 18 different places are shown in Table III.

TABLE III

Performance of senji No. 1 against local on zamindara lands

Place	Calculated yield in maunds per acre of		Percentage difference over control
	No. 1	Local	
Banga	320.0	256.0	25.0
Lalpur	185.6	154.9	7.5
Khanpur	164.2	146.2	13.8
Tabowal	264.0	216.0	22.2
Alawalpur	264.0	227.0	16.3
Najochak	125.2	84.1	46.2
Bhagowal	123.0	120.0	6.6
Amritsar	320.0	256.0	25.0
Dolo Nagar	200.0	140.0	42.8
Nawanshahar	371.3	318.8	16.3
Do.	270.0	255.0	6.0
Hundewala	252.0	250.0	..
Lutheri	223.1	200.5	11.8
Amsarali	137.0	177.5	5.0
Kangiwala	152.0	136.0	11.7
Chimi	152.0	150.8	..
Suchanlan	255.6	200.2	12.5
Gurdaspur	250.5	222.0	12.6

In these tests too, No. 1 showed marked superiority over the local and its increase in yield varied from almost equal to 46.2 per cent.

In view of its high forage yield at the Fodder Research Station, Departmental Farms and *zamindara* lands, superiority of No. 1 was established and recommended for cultivation by the *zamindars* in this State.

SUMMARY

Senji No. 1 is higher forage yielding variety than the local varieties but takes on an average 9-10 days more to flower and become ready for forage.

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A PRELIMINARY NOTE ON THE ESSENTIAL OIL BEARING PLANTS OF KASHMIR

(PART V)

By K. L. HANDA, L. D. KAPOOR and O. N. CHANNA, Drug Research Laboratory, Jammu

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IN continuation of previous communications (Parts 1, 2, 3 and 4) some more aromatic plants have been collected during the botanical tours of the different parts of Jammu and Kashmir State and results of preliminary study are given hereunder.

PINE NEEDLE OILS

The aromatic distillates obtained by the steam distillation of the needles and cones of various coniferous plants are called pine needle oils. These oils possess balsamic and refreshing pine-wood odour and, therefore, find extensive use in the preparation of various pine essences for spraying of dwellings and sick rooms, and in the preparation of aromatic baths, perfumery and finer soap industry. They are distilled from young leaves, twigs and one year old cones of pines, larches and firs. Quite a number of species of pines and firs grow wild in the forests of Jammu and Kashmir. These constitute perhaps one of the largest sources of national wealth and considerable revenue is realized by their exploitation for the country's timber requirements for building and other purposes.

The leaves of the various species of pines occurring in Jammu and Kashmir were procured in fresh condition and steam distilled.

Abies pindrow Royle-Himalayan Silver Fir (Vern. Tung)

It is an evergreen tree commonly growing wild at altitudes of 8,000 to 11,000 ft. It grows even above the deodar zone in the mountains bordering Kishanganga and in the forests of Sindh, Liddar and Jhelum valleys. The leaves have a very fine aroma. Collected from Gulmarg side the leaves on steam distillation yield a pale yellow oil with balsamic odour and with the following physical characteristics :

Yield of oil	0.61 per cent
Specific gravity at 15°C	0.8845
Refractive index at 20°C	1.4641
Solubility	It is soluble in 15 to 20 volumes of 80 per cent alcohol
Esters as bornyl acetate	30.8 per cent

Pinus longifolia Roxburgh (Vern. Chir)

This is an evergreen tree growing abundantly at altitudes of 2,500 ft. to 5,500 ft. in the Udhampur, Chenani, Reasi, Kathua forests division of Jammu Province. Large quantities of oleo-resins are tapped annually for the manufacture of terpentine oil and rosin at a factory situated near Jammu. Needles in fresh condition obtained from Udhampur forests on steam distillation yielded a pale yellow oil with the following characteristics :

Yield of oil	0.22 per cent
Specific gravity at 15°C	0.9116
Refractive index at 20°C	1.4845
Solubility	Soluble in all proportions of 90 per cent alcohol
Esters as bornyl acetate	15.4 per cent

Pinus excelsa Wall-Blue pine (Vern. Kail, Kairoo)

This is another evergreen tree which is very common in Kashmir forests at altitudes of 3,000 to 8,000 ft. It forms the major forest crop in Jammu and Kashmir. It is distributed in the valleys of Sindh, Jhelum, Krishanganga Lidder, Chenab, etc. The leaves of the tree procured from Gulmarg range were distilled and a pale yellow oil having balsamic odour and the following characteristics was obtained :

Yield of oil	0.38 per cent
Specific gravity at 15°C	0.9545
Refractive index at 20°C	1.4647
Solubility	Soluble in 13 to 20 volumes of 80 per cent and in all proportions of 90 per cent alcohol
Ester as bornyl acetate	23.3 per cent

Cedrus deodra Loud (Vern. Deodar)

This is an evergreen tree which is very common in Kashmir forests at altitudes of 5,000 to 10,000 ft. particularly in the Krishnaganga, Kishtwar and Jhelum valleys. It is exploited in large quantities for its valuable timber much appreciated and used for building purposes. The leaves possess very fragrant aroma. The leaves obtained from Gulmarg side on steam distillation yielded a pale yellow oil with the following characteristics :

Yield of oil	0.22 per cent
Specific gravity at 15°C	0.8645
Refractive index at 20°C	1.4721
Solubility	Soluble in 20-25 volumes of 80 per cent and in all proportions of 90 per cent alcohol

Picea morinda Link (Vern. Spruce, Kachhlu)

This is an evergreen tree growing at altitudes of 6,000 to 11,000 ft. in the forests of the State and is generally found in association with the broad-leaved trees or pine trees. It is common in Krishnaganga, Drawa and Sindh valley forests. The

leaves procured from Gulmarg range were steam distilled and a pale yellow oil with the following characteristics was obtained :

Yield of oil	0.22 per cent
Specific gravity at 15°C	0.9195
Refractive index at 20°C	1.4721
Solubility	Soluble in 20-25 parts of 80 per cent and in all proportions of 90 per cent alcohol
Esters as bornyl acetate	23.2 per cent

Taxus baccata Linn (Vern. Burmi)

The tree which is valued for its brick red wood is found in the pine forests of Jhelum, Kishanganga and Sindh valleys. On steam distillation only traces of oil were obtained from the leaves and, therefore, is considered to be of no practical utility.

Eucalyptus species

Eucalyptus trees are not indigenous to India but many species have been introduced from Australia in different parts of India notably in Nilgiris (South India) and oil of medicinal quality is being distilled there. Attempts to raise *Eucalyptus* were also made in northern India and *E. Viminalis* Labill and *E. citriodora* Hook were planted in Jammu many years ago. These have fared well under the local climatic conditions and have attained the normal height. The fresh leaves were steam distilled and a pale yellow oil was obtained from both the species. The characteristics of both the oils are recorded hereunder and compared with those of the Australian oils.

<i>Eucalyptus viminalis</i>	Local oil	Australian oil
Yield of oil	1.4 per cent	0.35 to 0.74 per cent
Specific gravity at 15°C	0.9007	0.044 to 0.9162
Refractive index at 20°C	1.476	1.4855
Solubility	Soluble in 2.3 volume of 80 per cent and all volumes of 90 per cent alcohol	In equal volume of 80 per cent alcohol
Percentage of cineol	17 per cent	50 per cent
Acid value	0.6	9.5
Ester value	16.0 Saponification value	
<i>Eucalyptus citriodora</i>	Local oil	Australian oil
Yield of oil	0.83 per cent	0.596 per cent
Specific gravity at 15°C	0.9207	0.864 per cent
Refractive index at 30°C	1.4924	1.4651 to 1.4678
Solubility	7-10 volumes of 80 per cent and 1 to 3 volumes 90 per cent alcohol	1.5 volume of 70 per cent alcohol
Saponification value	8.5	7.5

Ocimum species

There are several species of *ocimum* which grow wild and are cultivated in India. *Ocimum canum* grows wild in the areas neighbouring lower hills of Jammu while *O. basillicum* and *O. sanctum* are cultivated occasionally as garden plants in Jammu.

Ocimum canum (*Vern. Kala-tulsi*)—This is an erect and much branched herbaceous plant which grows all over India. It is extensively used in indigenous medicines for the treatment of fevers and skin diseases. The leaves and flowering tops of the plants growing wild in Jammu were procured and steam distilled. A sweet smelling oil was obtained with the following characteristics which are compared with those of the African oils :

<i>Ocimum Canum</i>	Local oil	African oil
Yield of oil from fresh leaves	0.5 per cent	0.65 per cent
Yield of oil from dry leaves	2.1 per cent	1.0330 at 32°C.
Specific gravity at 15°C	0.9105
Refractive index at 20°C	1.4908
Solubility	Soluble in 2 volumes of 80 per cent alcohol	2½ volume of 70 per cent and more of 75 per cent and in all proportions of 80 per cent alcohol
Acid value	2.7
Ester value	15.3	301

Ocimum basilicum Linn (Vern. Niazbaz)

This is an erect herbaceous plant which grows wild in the Punjab and is cultivated throughout the greater part of India. The leaves are extensively used in the indigenous medicine as carminatives, stimulant, demulcent and as febrifuge. The leaves and flowering tops of the plants raised in Jammu were steam distilled. A sweet smelling oil with the following properties was obtained :

Yield of oil from fresh leaves	0.53 per cent
Yield of oil from dry leaves	2.3 per cent
Specific gravity at 15°C	0.964
Refractive index at 20°C	1.505
Acid value	0.7
Ester value	21.9
Percentage of linalylacetate	3.6
Alcohol content with reference to linalool	17.7 per cent

Ocimum sanctum Linn (Vern. Tulsi)

This herb is cultivated throughout India near about sacred places. In the indigenous medicine the leaves are used as expectorant, stomachic, carminative, diuretic and in febrile conditions. Its leaves and flowering tops collected from the

cultivated plants in Jammu were steam distilled. The oil obtained had the following characteristics which have been compared with those of the foreign oils.

Ocimum sanctum	Local oil	Foreign oil
Yield of oil	0.9 per cent	0.6 per cent
Specific gravity at 15°C	0.967	0.952
Refractive index at 20°C	1.5197	1.5170
Solubility	Soluble in all proportions of 90 per cent alcohol
Saponification value	86	2.8

Mentha viridis Linn (Vern. Pudina)

Mentha viridis is extensively cultivated in India as a garden plant for household purposes. The green leaves are used in *chutneys* and other carminative household preparations. The fresh leaves and flowering tops were procured and steam distilled. A pale yellow oil with the following characteristics was obtained, these are compared with those of the American and German oils.

<i>Mentha viridis</i>	Local oil	American and German oil
Yield of oil	0.25 per cent	0.92 to 0.94
Specific gravity	0.94 at 18°C	1.482 to 1.489
Refractive index	0.539 at 18°C	
Solubility	1 to 3 volume of 80 per cent alcohol	1 to 1.5 volume and more of 80 per cent alcohol
Acid value	0.3	upto 2
Ester value	11.4	18 to 36
Percentage of carvone	20	42 to 60

Cinnamomum camphora Linn. Nees and Enerm (Lauraceae) Camphor tree, (Vern. Kapur, Karpuram)

Camphor tree is not indigenous to India but has been introduced for cultivation at a number of places in India. The plants have been experimentally cultivated in Dehra Dun, Lucknow, Mysore, Nilgiris, Madras, Cochin, etc. The trees did well in these places but the yield of the oil and camphor contents varied so widely that commercial exploitation was not economic. In Nilgiris this plant is exploited for commercial purpose to some extent.

Cinamomum camphora was also raised in Jammu (900 ft.) many years ago. The tree has done very well and attained normal height. Green leaves of this tree were collected in Spring and steam distilled. A pale yellow oil with camphor-like odour was obtained with following characteristics:

Yield of oil from fresh leaves	1.4 per cent
Specific gravity at 15°C	0.9117
Refractive index at 20°C	1.473

When the oil was chilled no camphor, however, could be separated.

Magnolia grandiflora Linn. (*Great flower Magnolia*, *Great Laurel Magnolia*)

This is an evergreen tree attaining a height of 80 to 100 ft. with firm laurel-like leaves and large whitish highly fragrant cup-like flowers. It is a native of South United States. It has been introduced in India and is cultivated for ornamental purposes in gardens all over the world including India.

The trees have been raised in Jammu. The flowers collected from local trees were steam distilled and yielded 0.12 per cent essential oil with very pleasant aroma characteristic of the flowers. It gave a refractive index 1.478 at 20°C. The other constants could not be observed as the oil was insufficient for study.

The following plants growing in Jammu and Kashmir have also been studied with regard to their essential oil contents but only traces of oil could be obtained from these plants.

Capparis spinosa Lamk. (Capparidaceae), *Caucalis latifolia* Linn. (Umbelliferae), *Senecio nudicaulis* Linn.-(Compositae), *Pimpemila diversifolia* Wall DC.(Umbelliferae), *Heracleum Cachminicum* Clarke (Umbelliferae), *Nepeta erecta*-Benth (Labiatae).

SUMMARY

Thirteen more essential oil bearing plants, both those growing wild and exotics introduced for cultivation, have been studied. The yield and the physical properties of these have been studied and compared with those growing in the foreign countries.

ACKNOWLEDGMENT

We are very grateful to Col. Sir R. N. Chopra for his kind guidance during the course of this investigation.

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RAPID ESTIMATION OF YIELD AND IODINE VALUE OF OILS IN SMALL SAMPLES OF OILSEEDS

By A. R. S. KARTHA, A. S. SETHI and K. C. GULATI, Indian Agricultural Research Institute, New Delhi

(Received for publication on 6 April 1954)

DETERMINATION of yield and iodine value of oil in small samples of seeds, using inexpensive methods, is of considerable importance for plant breeders. Methods available for rapid determination of oil in small samples of seeds were discussed in a previous communication from this laboratory [Chopra, *et al.*, 1952]. A rapid technique is now described to determine simultaneously both yield and iodine value of oil, using 0.3 to 0.5 gm. specimen of seed [Sethi, *et al.*, 1953].

If the iodine value of oils extracted with non-chlorinated solvents is to be determined, last traces of solvent must be removed whether the oil is to be weighed as such for the determination, or not. Sulphuric ether can be readily removed, but extraction with this solvent requires long time and is very expensive. When higher boiling solvents like benzene, petroleum ether b.p. 70-80°C, etc. are used, removal of last traces of solvent from extracted oil is tedious and inconvenient, particularly when only small specimens of seeds are available, this may also introduce error due to oxidation of oil exposed in thin films at higher temperatures, if due precautions are not taken. If extraction of oil from seed can be satisfactorily carried out with carbon tetrachloride or chloroform instead of petroleum ether or benzene which are employed when the oil content alone is to be determined

TABLE I
Extraction of oilseeds with benzene and with carbon tetrachloride

No.	Name of seed	Per cent oil yield	
		with benzene	with carbon tetrachloride
1	Linseed	46.1	46.6
2	Safflower seed	28.9	28.7
3	<i>Brassica campestris</i>	43.2	43.4
4	Tobacco seed	40.7	41.4
5	Poppy seed	48.9	50.1
6	Linseed R.R. 10	44.7	45.3
7	Groundnut	46.2	45.4
8	Mustard seed	35.4	35.4
9	<i>Til</i> seed	51.2	50.7
10	Linseed N.P. 12	40.1	42.5
11	Linseed R.R. 235	44.8	46.2

[Chopra, *et al.*, 1952], then an aliquot part of the extract can be used for determining the oil yield while other aliquots may be directly used for iodine value determination. This will be possible if the carbon tetrachloride or chloroform extracts all the oil from the seed, but does not extract any other material to any appreciable extent, and for this it has to be shown (a) that percentage of oil yield is the same in the case of carbon tetrachloride or chloroform as with benzene, and (b) that iodine value of oil extracted and determined by the suggested procedure agrees with that of the expressed oil determined by conventional methods. No quantitative investigations of this type have been recorded so far.

To demonstrate (a), 0.3 to 0.5 gm. specimens of 11 different oilseeds are extracted with benzene and with carbon tetrachloride. The weighed samples are ground with 2.5 gm. of anhydrous sodium sulphate and 2 gm. glass powder (pyrex glass, washed with hydrochloric acid) in a mortar at room temperature, transferred to a 50 c.c. Erlenmeyer flask with 10 c.c. of solvent, refluxed for 2-3 minutes, cooled to room temperature, and then filtered through a sintered glass funnel into a 25 c.c. graduated flask. The residue and flask are washed repeatedly with small amounts of cold solvent till the total filtrate is about 20-22 c.c. The solution is then made up to volume, and an aliquot part, usually 5 c.c., is evaporated in flat dish in a vacuum oven at 100° C. for one hour to obtain the oil yield. The results obtained with benzene show good agreement with those obtained with carbon tetrachloride (Table I).

To determine the iodine value using small quantities of oil two procedures are available: (i) the conventional method using reduced quantities of oil and reagents

TABLE II

Iodine values of some oils by the gravimetric bromine absorption method.

The groundnut, linseed, safflower seed and sesame oils used showed iodine values of 86.88, 174.175, 137.138 and 103.109 respectively by Hanus' method.

Oil	Container crucible		Container dish	
	Weight of oil mg. approx.	Iodine value	Weight of oil mg. approx.	Iodine value
Groundnut oil	9	81	9	77
	19	81	27	73
	29	84	24	72
	25	87	25	70
	49	90	25	72
	73	89	49	73
	98	88	71	74
	91	80

TABLE II—(contd.)

Iodine values of some oils by the gravimetric bromine absorption method

Oil	Container crucible		Container dish	
	Weight of oil mg. approx.	Iodine value.	Weight of oil mg. approx.	Iodine value.
Linseed oil	11	75	11	70
	21	101	21	85
	31	101	26	95
	40	109	31	81
	65	107	41	86
	87	106	51	96
	75	114
	97	113
Safflower seed oil	9	99	9	96
	18	117	19	94
	22	124	28	98
	27	127	36	105
	35	131	46	117
	44	132	91	132
	66	140
	88	140
Sesame oil	45	109		
	48	113		
	116	114		

and (ii) bromine absorption methods in gravimetric or volumetric forms. The volumetric form does not offer any advantages over the conventional iodine value methods using Wij's or Hanus' solutions. A number of papers have recently appeared advocating gravimetric bromine absorption methods for small quantities of oil and attempts were made to see whether the method could be adapted for use

in the present connection. For this a known volume of carbon tetrachloride solution containing approximately the desired quantity of the oil is pipetted into a clean, dry, weighed porcelain crucible or dish, solvent evaporated in a vacuum oven at 100°C for one hour, cooled and weighed to determine the exact weight of oil taken, and then exposed to the action of excess bromine vapour under a bell jar, in the dark, for one hour. Excess bromine is then removed by heating the container in a vacuum oven, in the dark, for one hour at 100°C or longer till constant weight is reached. Two types of vessels were used : porcelain crucibles having top diameter 2 inches, bottom diameter $\frac{3}{4}$ inch, and height $\frac{1}{2}$ inch, or flat porcelain dishes of height $\frac{1}{2}$ inch and diameter 3 and $\frac{1}{4}$ inches. The results obtained with different oils, mean of triplicates or quadruplicates (TABLE II) indicate the following :

- (i) Using same container, apparent iodine value tends to increase as the weight of oil taken is increased from about 10 to 100 mg.
- (ii) When small weights of oil are taken (10-100 mg.) apparent iodine values are smaller with containers of larger surface area (in this instance the flat dishes) for same weight of oil. This points to some dependence of apparent iodine values on comparative surface area of oil film.
- (iii) Apparent iodine values differ to varying extents with different oils depending on their component acid composition. The divergences are largest with linseed oil, showing that linolenic acid reacts in a characteristic way. The differences are smaller with oils which do not contain linolenic acid. With groundnut oil which contains mainly oleic as the unsaturated acid, apparent iodine value shows the nearest approach to normal.

Gravimetric bromine absorption methods under the experimental conditions described above cannot, hence, be used for accurate determination of iodine value of oils.

Conventional methods using reduced quantities of reagents and oil gave, with quantities of oil as small as 15-20 mg., values agreeing closely with that obtained with the normal procedure using 150-200 mg. of oil. That the oil extracted with carbon tetrachloride shows the same iodine value when determined by the present procedure, as that of the oil expressed and examined by the conventional method was shown as follows :

The oil was expressed from all the samples of seeds using a laboratory hydraulic press, filtered, and the iodine value determined by the conventional procedure (Hanus' method). Iodine value of the carbon tetrachloride extracted oil was determined by the following procedure : 0.3 to 0.5 gm. of seed was extracted with carbon tetrachloride, extract made up to 25 c.c. and 5 c.c. of this evaporated in a dish as already described : this gives the weight of oil in 5 c.c. of the solution. Other 5 c.c. aliquots were pipetted into iodine value flasks, 10 c.c. of Hanus' solution added and the determination conducted as usual, the only alteration being use of N/20 instead of N/10 thiosulphate solution. The results obtained (TABLE III) show that iodine values

TABLE III

Iodine values of expressed and carbon tetrachloride extracted oils

No.	Name of seed	Iodine value of expressed oil, conventional method	Iodine value of extracted oil, present procedure
1	Linseed*	183.9	182.3
2	Safflower seed	136.7	136.6
3	<i>Brassica campestris</i>	103.8	100.6
4	Tobacco seed	131.5	131.2
5	Poppy seed	130.7	130.2
6	Linseed R.R. 10*	174.5	173.6
7	Groundnut	85.5	88.8
8	Mustard seed	113.9	113.8
9	<i>Til</i> seed	108.0	108.5
10	Linseed N.P. 12*	163.0	164.5
11	Linseed R.R. 235*	178.4	177.0

*In case of the linseed oils the time of reaction using the present method had to be increased to two hours instead of the usual 30 minutes.

obtained by the present procedure, using small samples of seeds, agree well with those obtained by conventional methods on the oils expressed from the seeds. Using the present method, the yield and iodine value of the oil in a specimen of seed can be determined in about 1.5-2 hours.

SUMMARY

A method is described whereby the yield and iodine value of oil in a 0.3-0.5 gm. specimen of seed can be determined simultaneously in about 2 hours. The method is based on the use of carbon tetrachloride as solvent. Yield of oil is determined gravimetrically and iodine value by Hanus' method.

Determination of iodine values of small specimens of oil (10-100 mg.) by gravimetric bromine vapour absorption method showed that this technique is not quite suitable for determination of iodine value of oils and fats, especially the oils of higher unsaturation, under the experimental conditions studied.

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METHODS OF SURVEYING AND MEASURING VEGETATION

BY DOROTHY BROWN

(*Bulletin No. 42 of the Commonwealth Bureau of Pastures and Field Crops, Hurley, Berks. Published by the Commonwealth Agricultural Bureau, England, 1954, pp. XV and 223 with 45 figures and 46 tables, Price 35s.*)

THIS bulletin, as claimed by the author, is a review of the world literature on the methods used in quantitative ecological studies of grazing lands. There is no attempt of their critical evaluation. However, appraisal of values is frequently indicated in the text and it has been rightly left to the investigator to test the validity and usefulness of the methods in his own fields. An up-to-date bibliography given at the end will be found very helpful for the purpose.

The book is divided into four parts. Part I deals with vegetation and its sampling. Open and dense grasslands without and with varying density of shrubs are described in relation to height and habit of the grasses. This gives an easy physiognomic comprehension of the vegetation for which the methods are mainly designed. A very useful chapter on the theory of sampling by G.M. Jolly is added. It gives a clear understanding of the relation between sampling and aggregate errors. It is advantageous to learn at the outset the sources and nature of bias towards sampling and reduce it to the minimum. In the third Chapter is given the sampling apparatus which describes areas, quadrats, lines, points and enclosure and exclosure techniques.

The actual methods are described under three phases of vegetational survey, namely botanical analysis, productivity and utilisation each forming a separate section of the book. Techniques of estimating frequency, number, area and weight of the species are given in four chapters of Part II. Much information has been collected after a thorough probe into the literature which is not easily available to an average worker. It is interesting to note how ecologists have differed from each other regarding the relative importance of these distributional attributes of species.

Part III on productivity proceeds with the methods for measuring yield and grazing capacity with an evaluation of range condition. This is followed by a chapter each on estimate of cover, weight, area and weight, actual weight, the animal as a measure of pasture production, gross and net yields compared and pasture recording and regional surveys. The methods described in detail are objective and suited to all types of pasture. Their value in planning and research needs no emphasis.

Utilization methods are dealt in Part IV. The problem of utilization takes into consideration effective range management. Assessment of range condition and diagnosis of range deterioration are important features of this phase of study.

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